2019

Habitat Associations and Community Interactions of Non-Native Species in the Southern Basin of Lake Michigan

Erin O'shaughnessey

LOYOLA UNIVERSITY CHICAGO

HABITAT ASSOCIATIONS AND COMMUNITY INTERACTIONS OF NON-NATIVE SPECIES IN THE SOUTHERN BASIN OF LAKE MICHIGAN

A THESIS SUBMITTED TO

THE FACULTY OF THE GRADUATE SCHOOL

IN CANDIDACY FOR THE DEGREE OF

MASTER OF SCIENCE

PROGRAM IN BIOLOGY

BY

ERIN O’SHAUGHNESSEY

CHICAGO, IL

MAY 2019

ACKNOWLEDGMENTS

This research was funded by Illinois Department of Natural Resources. Thank you to my advisor Dr. Reuben Keller and my committee members Dr. Martin Berg and Dr. Tim Hoellein for their time and comments on my thesis. Thanks to Dr. Victoria Prescott for the many hours spent helping with data analysis and R, and at happy hours. Thanks also to Jenny Par and Deirdre Turner at Loyola University Chicago for their help in the lab and identifying mollusk samples. Thank you to Rachel Egly, Trent Henry, Jon Brenner, John Zink, Gabrielle Habeeb, Bridie Hulsebsoch, and Shehla Chowdhury for their help in the field and lab. Finally, thanks to my family and friends for their support during field work and the writing process.

To my parents and brothers for their support in all endeavors.
In memory of Dave Shaughnessy, who passed away while assisting with this research.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Non-native crayfish, mollusks, and macrophytes can have large impacts on biodiversity and damage ecosystem services in freshwaters. In 2015 we discovered an established population of the globally widespread invader red swamp crayfish (*Procambarus clarkii*) in the North Shore Channel of the Chicago Area Waterway System. This population overlaps with a population of rusty crayfish (*Faxonius rusticus*), a previous invader that is widely distributed and usually the dominant crayfish species across the Great Lakes region. I studied the interactions between these two species while directly competing over shelter and food. In the field, each species was studied to determine the rate of predation in a channel with murky water, and a harbor with clear water. The overlapping population of crayfish in the North Shore was discovered while sampling for crayfish species throughout the Chicago region. Crayfish are poorly sampled in the Great Lakes Region, leaving large gaps in knowledge of native and non-native crayfish distributions. I examined the role that artificial habitat and anthropogenic changes have on crayfish distribution and created an updated distribution of crayfishes in the Chicago region. I also studied the relationship between the arrangement of anthropogenic habitat and non-burrowing mollusks and macrophytes. The density and species composition of mollusks and macrophytes were compared between habitat types. I aimed to study the role that human habitats play in the distribution of non-native and native crayfish, mollusks, and macrophytes.

CHAPTER I

INTRODUCTION

Non-Native Species

Freshwater ecosystems have high biodiversity and provide vital ecosystem services, making them essential to human communities around the world. Some of the main human uses of freshwater ecosystems include transportation, drinking water, and irrigation of crops. The importance of these services means that freshwater ecosystems are regularly accessed by humans, which leads to a high likelihood that new species will be transported to freshwaters where they were not previously found. The high biodiversity of freshwater systems results in a higher potential for biodiversity loss and damage to ecosystem function if a non-native species becomes established in an area (Ricciardi & Rasmussen 1999; Havel et al., 2015). Non-native species can cause immense ecological and financial damage when they enter new ecosystems (Mills et al. 2004; Bax 2003). Ecologically, invasive species can alter food webs in many ways, including by decreasing the food source of other organisms, predation, and the introduction of new diseases (Johnson et al. 2009; Kreps et al. 2016). They can also change the physical characteristics of a habitat through, for example, increased turbidity or altered stream beds (Albertson and Daniels 2016). These impacts can multiply, for example altered stream beds potentially alter the ability of macrophytes to grow, further disrupting food webs (Kreps et al. 2016). Financially, invasive species can cause large impacts including clogging pipes, damaging populations of commercially or recreationally important fishes, and increasing erosion of streams (Pimentel 2005; Pejchar and Mooney 2009; Oreska and Aldridge 2011; Keller et al. 2018).

Invasive species are found in every ecosystem type around the world, including marine, freshwater, and terrestrial. In the United States, a species is typically considered non-native if it was not present prior to European colonization (Ricciardi 2006). The invasion process begins when a species is moved from one area to another (Figure 1). Throughout this thesis, a species is considered *introduced* when one or more individuals are transported to, and are then released or escape into, an area where the species was not found prior to European colonization. Species that then establish reproducing populations are referred to as *established*, and several factors are known to increase the chance of establishment. These factors include a lack of predators in the new environment, high competitive ability of the new species, and high reproductive potential of the non-native (Mills et al. 2004). Finally, established species that cause harm or have the potential to cause harm to native biota, human health, and/or the environment, are referred to as *invasive* (Figure 1). Although the term *invasive* is sometimes applied to any non-native organism, throughout this thesis I use the 'harm' definition, which is used by U.S. federal agencies (Clinton 1998).

1) Non-native: Species is entrained in vector and moved beyond its native range

    Species does not survive transport

2) Introduced: Species survives transport and release

    Species fails to establish

3) Established: species begins reproducing beyond human cultivation

    Species does not cause harm

4) Invasive: species spreads and causes harm

Figure 1. Steps in the invasion process (adapted from Kolar & Lodge (2001)). Green arrows indicate the successful movement of a species through steps of the invasion process. Red arrows indicate a species failing to move through the process.

Freshwater Invasions

Freshwater ecosystems have higher biodiversity per surface area when compared to terrestrial and marine ecosystems (Dudgeon et al. 2005). In freshwaters, invasive species often outcompete native species to become established. Due to their higher biodiversity, invasions in freshwaters are generally more damaging to biodiversity than invasions in marine and terrestrial systems (Dudgeon et al. 2005; Ricciardi & Rasmussen 1999). Freshwater systems play a large role in nutrient and water cycling, making them important for the supply of ecosystem goods and services (Havel et al. 2015). Invasive species, as well as land use changes, have put freshwater systems such as lakes and streams at high risk of damage (Ricciardi & Rasmussen 1999). Impacts of invasive species include declines in populations of native species, changes in physical ecosystem structure, and changes in nutrient cycling (Bernt et al. 2014; Ricciardi et al. 2011).

Humans have facilitated the spread of aquatic invasive species (AIS) within freshwater systems directly and indirectly by creating a range of vectors and pathways for their movement (Keller et al. 2011). For example, artificial canals often connect freshwater ecosystems that were previously disconnected and thus allow for travel of non-native species to new areas. Other vectors include increased shipping and the associated movement of ballast water, trade in live organisms, and movement of species for stocking and aquaculture (Havel et al., 2015; Ricciardi et al. 2011). These vectors allow non-native species to be accidentally or intentionally introduced into new ecosystems, and many of these species become invasive (Havel et al., 2015).

Presence of invasive species cause native species' decline in abundance and diversity in freshwater species including macrophytes, zooplankton, crayfishes, and fish (Hermoso et al. 2011; Gallardo et al. 2016; Lodge 2000; Harvey et al. 2011). For example, lakes invaded by common carp (*Cyrinus carpio*) have a lower abundance of native fishes (Weber and Brown

2011). These lakes also have altered habitat, nutrient availability, and macroinvertebrate communities (Weber and Brown 2011). Common carp physically alter the ecosystems they invade through their feeding behavior, which increases water turbidity (Drenner et al. 1997).

Zebra mussels (*Dreissena polymorpha*) and quagga mussels (*Dreissena bugensis*) are bivalves that invaded Europe and North America and caused extensive economic and ecological damage (Drake & Bossenbroek 2004). These organisms can reach population densities up to 500,000 individuals per square meter (Martel and Madill 2018). Zebra and quagga mussel populations clog water intake pipes which can impact water access for municipal and industrial users (MacIsaac 1996). This can cause economic losses due to the efforts involved in clearing the pipes. Zebra mussels can increase water clarity which increases light transmittance and increased growth of algae and benthic plants (MasIsaac 1996). The desire to avoid these and other impacts has made preventing the arrival and establishment of new freshwater non-native species a management priority across the globe.

Due to their role as ecosystem engineers and ability to physically alter ecosystems, invasive crayfishes are of concern (Statzner et al 2000). Invasive crayfish disrupt the ecosystem biologically and physically, sometimes also causing financial impacts, by damaging macroinvertebrate and fish communities (Lodge et al. 1994; Pimental 2005; Pejchar and Mooney 2009; Oreska and Aldridge 2011; Wilson et al. 2004). This reduces food supply for other predators of macroinvertebrates and can have food web impacts that reduce predator and sport-fish populations (Kreps et al. 2016). The reduction in fish populations can decrease food stability by damaging the recreational and commercial fishing industries (Pimentel 2005; Pejchar and Mooney 2009*).* Invasive crayfishes such as the rusty (*Faxonius rusticus*) and signal crayfish (*Pacifastacus leniusculus*) alter their physical habitat via feeding, burrowing, and fighting

(Harvey et al. 2001; Albertson and Daniels 2016). These behaviors increase water turbidity, can reduce fine sediment accumulation stream bed stability and increase erosion (Albertson and Daniels 2016; Harvey et al. 2001; Rodríguez et al., 2003; Souty-Grosset et al. 2016).

Two non-native crayfish species with particularly large and well-researched impacts are the red swamp (*Procambarus clarkii*) and rusty crayfish (*Faxonius rusticus*). Rusty crayfish have become the most widespread crayfish in the Midwest and have reduced native crayfish population in areas in which it has invaded (Lodge et al. 1994; Hein et al. 2006). This species has also been introduced to the Western United States and throughout the world (Peters et al. 2014). Red swamp crayfish has become a worldwide invader, causing impacts to native aquatic organisms and to human food security (Cruz and Rebelo 2006; Yue et al. 2010). This species has been found to reduce food security by eating rice seedlings in Asia, decreasing crop production and damaging irrigation systems (Yue et al. 2010). Additionally, it has been introduced to Eastern Africa, where there are not any native crayfish species (Nunes et al. 2017).

The state of Illinois contains a large network of freshwater ecosystems, many of which are connected to other states through rivers and lakes (Figure 2). Most of the state of Illinois is within the Mississippi River catchment. (Figure 2). The only region outside of this catchment is the northeast corner of the state which borders Lake Michigan, one of the Laurentian Great Lakes (Figure 3). Lake Michigan is also bordered by Indiana, Michigan, and Wisconsin, making it important to many different ecosystems and human communities. Through the St. Lawrence River, the Laurentian Great Lakes eventually reach the Atlantic Ocean. Throughout the modern history of North America, the Great Lakes and Mississippi River Basin have been an important resource for transportation and trading of goods. It is estimated that over 50,000 non-native species have been introduced to the United States (Pimentel 2002), with 10% of those becoming established, and only 10% of established species are estimated to become invasive (Groves 1991).

Figure 2. Map of Mississippi River Basin and Great Lakes Basin.

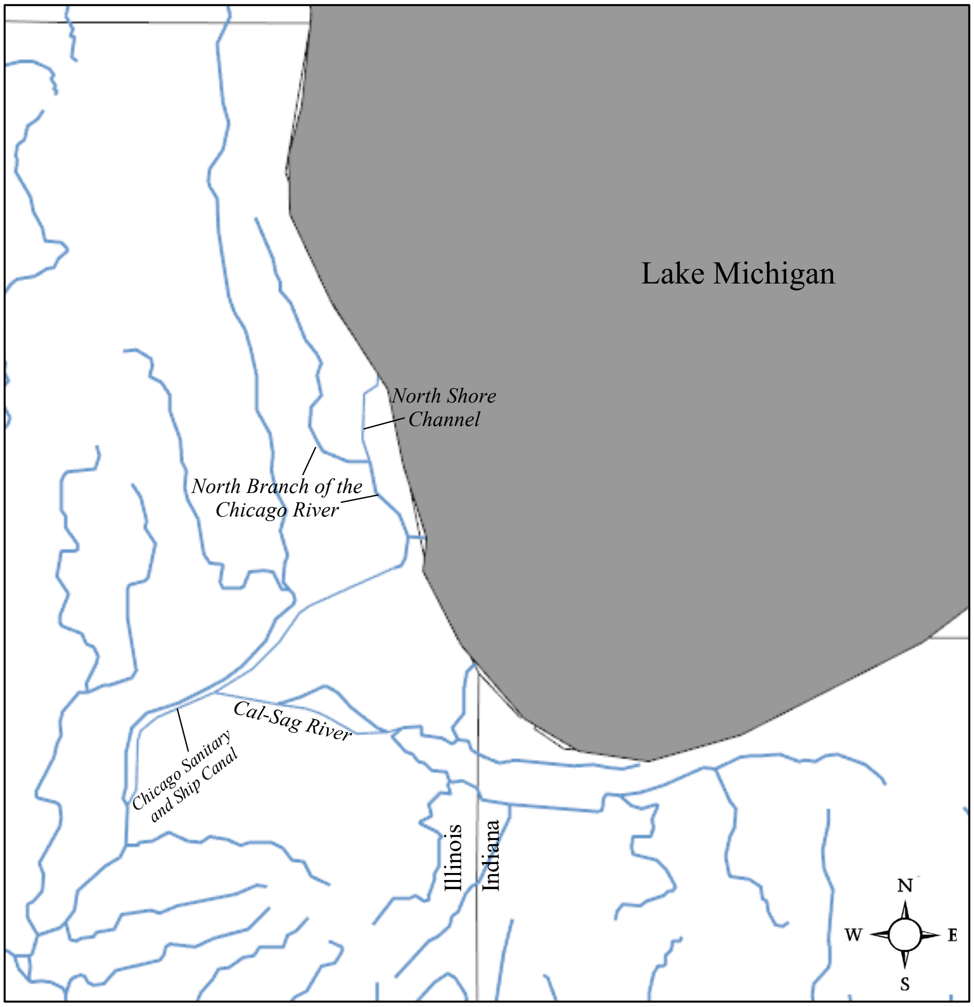


Figure 3. Map of north-east Illinois and north-west Indiana waterways.

Study Area

Chicago is a large city located on Lake Michigan, which straddles the boundary between the Great Lakes and Mississippi River basins. In 1900 the Chicago Sanitary and Ship Canal was opened, connecting these two basins with a permanent waterway (Hill 2000). The construction of this canal allowed the flow of the Chicago River to be reversed, taking waste and storm water away from Chicago and importantly protecting Lake Michigan – from which Chicago draws its drinking water – from this sewage. It also became an important waterway for shipping. In 1900, the Calumet-Sauganashkee (Cal-Sag) Channel was opened, connecting the Calumet River and the Chicago Sanitary and Ship Canal (CSSC), and in 1910 the North Shore Channel (referred to as the Channel from here on) was completed (Hill 2000). This Channel connects Lake Michigan at Wilmette Harbor to the North Branch of the Chicago River, and was constructed to increase water flow in the River and dilute effluent from the O'Brien Wastewater Treatment Plant. These waterways, and the rivers they are connected to, now form the Chicago Area Waterway System (CAWS) and continue to function in the same ways, remaining important for removal of storm water and wastewater, as well as for commercial and recreational navigation. Nearly half of the CAWS is man-made, with the remainder being highly altered natural watercourses (Hill 2000). This addition of pathways potentially allows for organisms to travel between the Great Lakes and Mississippi River Basins more easily, increasing the chances for a non-native species to reach a new area.

Chicago is a likely hotspot for future aquatic invasions for four main reasons. First, it has a large human population consisting of 2.7 million people in the City of Chicago, and over nine million people including the surrounding suburbs (US Census Bureau). This large population size increases the potential for introduction of non-native species from the pet trade, food industry, or accidental introductions (Havel et al. 2015). Additionally, the substantial amount of artificial and altered habitat in the Chicago region may allow for introduced species to become established, due to increased hard substrate and habitat changes. Included in this artificial habitat is the creation of harbors along the shoreline.

Chicago's role as a transportation hub is the second reason as to why we expect that this region is a potential hotspot for new invaders. Thousands of boats are docked in the Chicago region and in Illinois, boaters travel between multiple lakes and rivers throughout the region every summer (Cole et al. 2019). Recreational boating is a potential vector for transportation of non-native species between waterbodies. Along with recreational boaters, there is a large amount of commercial boating that occurs throughout the Chicago Region. Connections to the Mississippi River Basin allow species to be transported north into Chicago from the southern United States (Figure 2). Additionally, species can be transported from the Great Lakes Basin into the Mississippi River Basin. Throughout the history of the United States, Chicago has played a vital role in the shipping industry because its harbors allowed for aquatic transportation and trade between the Eastern and Western United States. Calumet Lake and Harbor still play an important role in both national and international shipping. Boats from continental United States and international areas such Asia and Africa, regularly visit Chicago harbors and can deliver organisms in ballast water, as hull foulers, or as contaminants of cargo (Johnson et al. 2001; Karatayev et al. 2014)

Third, Chicago is connected by continuous waterways to areas as far west as Montana, as far north as Canada, as far east as the Atlantic Ocean, and as far south as New Orleans. The connection to the Mississippi River basin allows for organisms from those areas to move or be transported to the Chicago region. The Mississippi River creates a drainage basin of 1,245,000 square miles and reaches parts of 31 states within the United States (US EPA). This widespread connectivity allows for easier movement of aquatic non-native species through the waterbodies throughout the United States. The Great Lakes Basin, which connects to the Mississippi River basin through Chicago, contains 84% of North America’s freshwater and a human population of 30 million (US EPA). The large human population combined with its importance as a source of freshwater, makes it important to monitor the impact of invasive species. Chicago’s location bridging the Mississippi River Basin and the Great Lakes Basin results in a vast number of waterbodies that have aquatic access to the Chicago region.

Finally, Chicago's location in the Southern Basin of the Great Lakes means that it will see the effects of climate change sooner compared to elsewhere in the Great Lakes. Climate change can augment the invasion process (Bellard et al. 2013) because increasing temperatures and changing climates result in changes in ecosystems, potentially allowing non-native species to travel to new areas. Since 1990, the average air temperature of the Great Lakes region has risen by 1.1°C (GLISA 2014), and it is predicted to continue rising. From 1973 – 2010, the annual average ice cover in the region has decreased by 71% and the frequency and intensity of storms has increased (GLISA 2014). Increased water temperature, along with high nutrient levels , has led to an increase in harmful algal bloom occurrences in the Great Lakes, causing areas of hypoxia and negative health impacts to wildlife and humans (GLISA 2014). The effects of climate change will likely allow additional species to establish in this area due to increasing water temperatures (Bellard et al. 2013).

The U.S. Army Corps of Engineers (USACE) reports that 254 aquatic introduced species have been found in the Great Lakes Basin, the Mississippi River Basin, or both (GLMRIS 2014). Establishment of non-native species caused by deliberate release have declined over time but unintended releases have increased (Riccardi 2006). The list of non-native aquatic species created through the Great Lakes and Mississippi River Interbasin Study (GLMRIS 2014) includes 87 species that are considered to pose a high risk to move into the Mississippi River Basin from the Great Lakes, and 57 species that pose a risk to move from the Mississippi River Basin into the Great Lakes (GLMRIS 2014). Seven species are not found in either basin, but are found in bordering watersheds and have the potential to move into either basin (GLMRIS 2014). This list includes species of macrophytes, amphipods, and fishes. The report provides a useful resource for future sampling efforts that aim to monitor where and when these species spread.

Despite this, systematic sampling of organisms in the Chicago region has been limited to just fishes. The lack of sampling for other taxa represents a lost opportunity to monitor and manage the spread of non-native species, and to rapidly detect new arrivals. Early detection while populations are small gives the best chance for eradication prior to the spread of new invaders (Myers et al. 2000; Hoffman et al. 2015). If undetected, these new species could result in significant harm to waterways across the Great Lakes and Mississippi River Basins.

Thesis Project

In Illinois, taxa such as plants, mollusks, and crayfishes pose high risks of invasion, but their populations are not well sampled in Lake Michigan and the Chicago area. Models that predict the potential spread of non-native crayfish usually focus on physical factors (e.g., water quality, habitat availability, climate), however, interactions between species can also be important for spread (Crall et al. 2006; Fletcher 2007; Weis 2010; Behringer and Hart 2017). Non-native mollusks and macrophytes can damage physical aspects of aquatic habitats, damage native species populations, and cause immense financial damage (Karatayev et al. 2014; Havel 2015). Despite the immense potential damage caused by aquatic invasive species in Chicago, macrophytes, mollusks, and crayfish are largely unstudied.

The first study to confirm a reproducing population of *P. clarkii* in the Chicago area was conducted by the Keller Lab in 2015. Chapter II builds upon this discovery to better understand the extent of the population, its relationship with other crayfish in the system, and potential impacts. *P. clarkii* are native to the southern United States, as far north as southern Illinois, and have proven to be successful invaders in other areas (Gherardi et al. 2002). The species exhibits a short life cycle and high fecundity (Gherardi et al. 2002) and has been shown to be competitively dominant when it enters new communities (Yue et al. 2010). The species can live in extreme environments including polluted habitats and temporary streams (Gheradi et al. 2002; Scalici & Gherardi 2007). These traits make it a high risk in the Chicago area and the Great lakes because the chances of the population spreading are high, due to its high ability to become established in ecosystems. It is important to identify where populations have established to better guide management efforts in the Chicago area to prevent spread throughout the Great Lakes watershed. Chapter II examines the relationship between a new invader, the red swamp crayfish, and a previously established invader, the rusty crayfish. Organisms must compete for resources such as food and shelter and be able to successfully avoid predation; we used lab and field experiments to study which species was more competitive and was better able to avoid predation.

The distribution of crayfishes in the Chicago region has received very little attention, leaving large gaps in our knowledge of the species present and their ranges. We studied the current distribution of native and non-native crayfishes in the Chicago region and the potential habitat associations of these species and densities that they are found in. The goal of Chapter III was to assess crayfish species location throughout the Indiana and Illinois portions of Lake Michigan and nearby stream, river, and lake habitats. I classified the sampling sites into six categories: stream, river, inland lentic, shoreline, harbor, and offshore. Species distribution, crayfish densities, and crayfish size were compared between these habitat types. We then compared our sampling results from 2015-2018 to historical crayfish distribution in the Great Lakes region to examine range expansions of non-native crayfish species in the area.

The role of artificial and anthropogenically-altered habitats in the distribution of non-native crayfish, freshwater mollusks, and macrophytes is largely unstudied. We aimed to determine if the habitats along the Illinois portion of the Lake Michigan shoreline influenced the distribution of non-native mollusks and macrophyte species. In marine systems, harbors are known to act as hotspots for non-native mollusk species (Airoldi et al. 2015). Hard anthropogenically-created marine substrates such as piers and harbors are known to have higher occurrences of non-native invertebrate species compared to native species (Airoldi et al. 2015). This relationship between anthropogenic structures and non-native species distributions has not been studied in freshwater. Chapter IV examined the arrangement of anthropogenic habitats in the Illinois portion of Lake Michigan and the relationship with mollusk and macrophyte species distribution and diversity. Through intensive sampling, we aimed to determine the prevalence and distribution of invasive macrophytes, epifaunal bivalves, and snails in the region. Macrophyte diversity and density were compared between habitat types and to the occurrence of non-native aquatic plant species. Additionally, the role that anthropogenic habitats, such as harbors, play in the establishment of macrophytes species along the shoreline was studied. Snail and bivalve distributions were examined and compared to species diversity and habitat types throughout the Chicago region. My goal was to determine the role that human altered habitats play in the distribution of non-native aquatic species.

CHAPTER II

WHEN INVADERS COLLIDE: COMPETITION, AGRESSION, AND PREDATOR AFFECT OUTCOMES IN OVERLAPPNG POPULTIONS OF RED SWAMP (*PROCAMBARUS CLARKII*) *AND* RUSTY (*FAXONIUS RUSTICUS*) CRAYFISHES

Introduction

The spread of non-native invasive species is a globally important driver of ecosystem service and biodiversity loss. Freshwater ecosystems are often strongly impacted, with effects including reduced size of and access to fisheries, reduced water availability for irrigation and municipal use, impeded navigation, and increased habitat for vectors of human disease (Pimental 2005; Pejchar and Mooney 2009; Oreska and Aldridge 2011; Keller et al. 2018). Invasive freshwater crayfishes can have particularly large ecological and economic impacts. Crayfishes are the largest freshwater invertebrates and many act as ecosystem engineers. The negative impacts of these species include decreased water quality (Souty-Grosset et al. 2016), altered aquatic macroinvertebrate communities (Wilson et al. 2004), reductions in macrophyte biomass and biodiversity (Lodge et al. 1994; Wilson et al. 2004), and extirpation of native crayfish. Crayfish can also be vectors of disease, including the crayfish plague (*Aphanomyces astaci*) which has been an important agent in the displacement of native European crayfish species by the invasive North American species which are immune to the disease (Souty-Grosset et al. 2016; Donato et al. 2018).

Much research has been conducted to model the distribution and spatial patterns of non-native freshwater species. This work usually aims to predict future spread of invaders for management (Clarke Murray et al. 2014). Although models usually predict potential spread as a function of physical factors (e.g., water quality, habitat availability, climate), interactions between invaders and other species can also be important (Crall et al. 2006; Fletcher 2007; Weis 2010; Behringer and Hart 2017). In particular, previously established species may compete with or predate upon the new arrival, and this can mediate the habitats into which freshwater non-native species can spread (Weis 2010).

The establishment and spread of non-native freshwater crayfishes may be particularly dependent upon their interactions with existing crayfish. Crayfish communities are usually composed of just one or two species, and when non-native species arrive and spread they often come to dominate the resulting crayfish community. For example, the spread of rusty crayfish (*Faxonius rusticus*) across the U.S. Midwest is associated with massive declines in population sizes of the existing crayfish species (the native virile crayfish (*Faxonius virilis*), and the northern clear-water crayfish (*Faxonius propinquus)*, which was likely an earlier invader (Olden et al. 2006). *F. rusticus* are now the dominant crayfish in most Midwestern freshwater habitats in which they are established. Likewise, the invasion of Europe by the North American signal crayfish (*Pacifastacus leniusculus*) has caused the widespread decline in native species and communities are now dominated by the invader (Westman et al. 2002; Dunn et al. 2009). In each of these cases the invader has been shown to be competitively dominant for resources such as food and shelter, and this is presumed to be a main mechanism of success.

Models of crayfish spread which use only physical factors in the native range to predict potential future range often perform poorly and underestimate the total range into which a crayfish may spread (Larson and Olden 2012). This indicates that crayfish native ranges are constrained not just by their tolerances of physical factors, but also by their interactions with other species and their ability to access new habitats. In turn, this demonstrates that understanding the outcome of biotic interactions is important for modeling the potential spread of invasive crayfishes.

In 2015 we discovered established and overlapping populations of the invasive rusty crayfish (*Faxonius rusticus*) and red swamp crayfish (*Procambarus clarkii*) in the North Shore Channel of the Chicago Area Waterway System (USA). *F. rusticus* (previously *Orconectes rusticus*; Crandall and De Grave 2017), are abundant in the Laurentian Great Lakes region after spreading from the Ohio River drainage in the 1970's (Wilson et al. 2004; Peters and Lodge 2014). This species has displaced native crayfish species in multiple waterways throughout the U.S. Midwest (Butler and Stein 1985; Gherardi and Daniels 2004) and has large ecosystem impacts including the alteration of whole lake foodwebs and reductions in sport-fish populations (Kreps et al. 2016). *P. clarkii* (red swamp crayfish) is a relatively recent invader of the Great Lakes Region that is native to the southern United States as far north as southern Illinois (Taylor et al. 2015). Within the Great Lakes basin, it is known to have isolated populations in Illinois and Michigan. Established populations in Wisconsin have been eradicated (Wisconsin DNR). There is concern that this species will continue to spread in the Great Lakes region, with impacts similar to those seen in other regions where it is established (Donato et al. 2018). Although good sampling data prior to 2015 are not available, the invasion histories of these species make it reasonable to infer that *F. rusticus* was established in the North Shore Channel prior to the arrival of *P. clarkii*. To the best of our knowledge this is the first example of these two invaders having overlapping populations in an area where they're both deemed invasive.

Here, we have made field observations and conducted lab and field experiments to investigate the potential for competition and predation to affect the persistence and spread of these species. We sampled from the overlapping populations to determine size distributions of each species as this is often an indicator of competitive dominance (Rabeni 1985; Klocker and Strayer 2004). Based on sampling results we designed a series of lab experiments to test for dominance between *P. clarkii* and *F. rusticus* at accessing limited shelter and food. Sampling observations and experimental results suggested that *P. clarkii* are more aggressive and less likely to seek shelter when threatened. We hypothesized that this would expose them to greater predation pressure, and tested for this in a field experiment that covered different habitats. Our work shows that competitive dominance may be associated with higher risk of predation and shows that these interactions will likely be important mediators of future spread of these species.

Methods

Trapping

The North Shore Channel (hereafter: the Channel) is a slow moving canal that was constructed between 1907 and 1910 to connect the North Branch of the Chicago River to Lake Michigan (Figure 1). Its habitat is homogenous, with a maximum depth of 2.9 meters, a consistent width of ~20m, and almost entirely soft mucky substrate. The Channel connects to Wilmette Harbor in Lake Michigan at its north end, and to the North Branch of the Chicago River at its south end. It was constructed primarily to deliver water from Lake Michigan to the O'Brien Sewage Treatment plant which is located nearby to where the Channel meets the Chicago River. Additionally, during high-flow events the weir at Wilmette Harbor can be opened to allow water to flow into Lake Michigan, reducing flooding throughout the surrounding urban area.

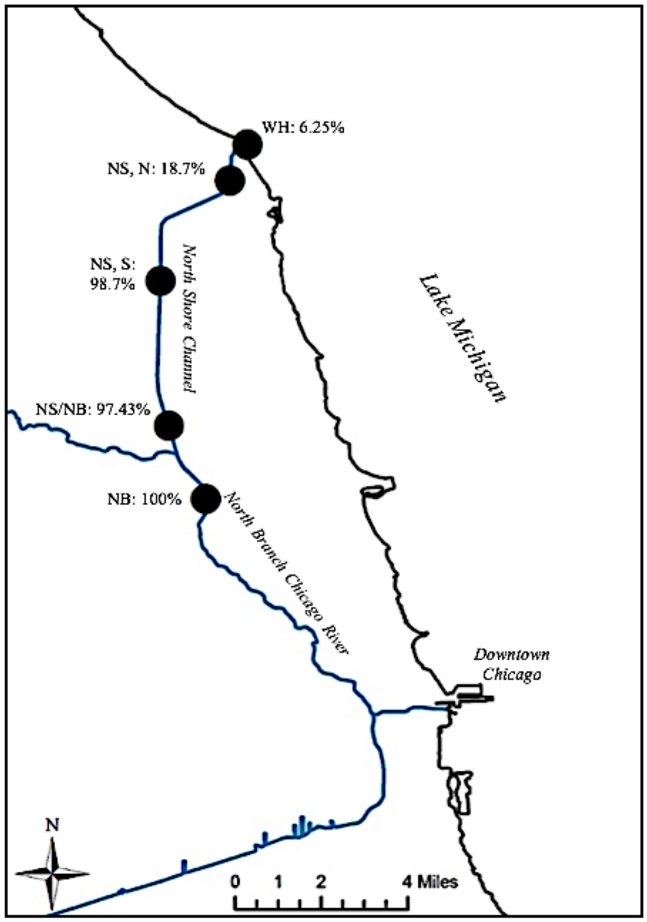


Figure 4. Map of crayfish sampling locations in the Chicago Area Waterway System. The percentage of *Procambarus clarkii* found at each site is given, and we note that all other crayfish were *Faxonius rusticus*. WH: Wilmette Harbor; NSN: North Shore Channel North; NSS: North Shore Channel South; NS/NB: North Shore Channel/North Branch; and NB: North Branch of the Chicago River

Crayfish populations in the North Shore Channel, North Branch of the Chicago River, and Wilmette Harbor were surveyed during 2015 at five locations (Figure 1). Subsequent sampling was conducted during summers 2016 and 2017. In all cases sampling was conducted using standard minnow traps baited with dry dog food. Traps were modified by increasing the openings to ~5cm diameter (Capelli and Magnuson 1983). Crayfish are most active at night, and traps were set one day and recovered the next. We recorded species, sex, and carapace length (CL; the length from the tip of the rostrum to the posterior end of the carapace) for all crayfish sampled. Crayfish were either released back where caught, or taken to Loyola University Chicago for experimental purposes.

A first observation was that we only found the non-native species rusty (*Faxonius rusticus*) and red swamp crayfish (*Procambarus clarkii*) in the Channel. Populations of these species overlapped, and multiple size classes of both species were found over all three years indicating that they are each well established (see below for full sampling results). These observations motivated the competition and predation experiments described in the following.

Competition experiments

To investigate competition between *F. rusticus* and *P. clarkii* for food and shelter, we conducted lab experiments at Loyola University Chicago during August – October 2016. All crayfish used in the experiments were collected during August and October of 2016 from the overlapping populations in the North Shore Channel. All collected crayfish were kept in large cattle tanks for at least one week to acclimatize to lab conditions. Individuals were not re-used for any experiments.

Shelter – Two Species

Shelter competition experiments were used to test which species is dominant for accessing a single shelter when individuals of both species are threatened. These experiments were conducted in ten-gallon aquaria at Loyola University Chicago. Aquaria were located in a closed lab with no activity from humans (apart from what was necessary to simulate fish attacks, see below) visible to the crayfish. Each aquaria was filled with ~8 liters of water, giving a depth of 15cm. Our methods followed those of Alonso and Martinez (2006), and a total of 23 trials were conducted.

For each trial, one of each species of crayfish was selected while ensuring that the carapace lengths of the two crayfish differed by no more than 10%. Crayfish were acclimated in the aquaria for 24 hours with a divider preventing physical interaction. Each crayfish was provided a 10-cm length of PVC pipe for shelter.

To begin the experiment the central divider and both shelters were removed from the tank and a single shelter (a 12cm long, 5.1 cm diameter piece of PVC pipe closed at one end) was added. Trials lasted 20 minutes, and at five, ten, and fifteen minutes we used a plastic fish to simulate an attack on the crayfish. Attacks included the fake fish directly interacting with each crayfish for an equal amount of time. Trials were recorded by video to minimize the potential for observers to affect behavior. Videos were later examined to determine the response of each crayfish to the attacks. Additionally, at 10 second increments, we recorded a) whether each species was in or out of the shelter; b) the behavior of each crayfish while out of shelter (active/passive); and c) the behavior of the crayfish in relation to each other. The behavior of the crayfish in relation to each other was quantified on a scale ranging from -2 to 5 (Table 1) following Karavanich and Atema (1998).

Food Competition

To examine competition between the two species for access to food we conducted 21 feeding trials using similar methods to Szela and Perry (2013). One individual of each species was haphazardly selected while ensuring that the size difference between the two crayfish was no more than 10% of CL. Pairs of crayfish were acclimated for 24 hours in 10 gallon aquaria filled to 15cm depth. No food was available for a minimum of 48 hours prior to each trial.

To begin the experiment, both crayfish were placed on one side of a divider and a 2cm piece of nightcrawler earthworm was placed at the opposite end of the tank. The divider was then removed. We recorded the first individual to access the food, the crayfish that ultimately ate the food, and the time elapsed before the food was fully consumed.

Predation Experiment

Predation experiments were conducted in the field to determine relative rates of predation on the two crayfish species in two different habitats. Experiments were conducted in the Channel and in Wilmette Harbor during June and July 2017 (see Figure 1 for locations) and involved tethering crayfish to weights, leaving them overnight, and checking the next day to see which had been removed. Methods followed those of DiDonato and Lodge (1993) and Childress and Herrnkind (1994). Crayfish were collected from the North Shore Channel and Wilmette Harbor using the sampling methods described above. All individuals used in these experiments were identified to species, measured for carapace length, and sex was determined.

To tether crayfish we cleaned the top of the carapace with 75% alcohol and used superglue to attach a small swivel. This swivel was tied to a 30cm long piece of four pound test strength monofilament fishing line, which was in turn tied to a hook in the center of a 15x15cm tile. To ensure that the tethers held, we included two controls of each species, at each site, each day. These individuals were glued in the same way but were placed inside minnow traps with the openings closed. None came free of their swivels or line.

Tiles were placed on the bottom of the habitat at least 5m apart and left overnight. In Wilmette Harbor the tiles were placed along the edge of the harbor wall. Tiles in the Channel were placed in ~1.5m of water along the bank, alternating species. In the Channel 63 *P. clarkii* and 37 *F. rusticus* trials were conducted. At Wilmette harbor, 60 *P. clarkii* and 41 *F. rusticus* trails were conducted, where each trial was a 24-hour period that an individual was tethered. Secchi depth was recorded at three points within each site daily. Tethered crayfish were checked every day for presence/absence and any that were missing were replaced by new crayfish. Missing crayfish were considered to have been predated. This assumption was supported by the controls described above, and by us frequently finding torn pieces of crayfish still attached to swivels.

Chi-square with Yates correction, t-test, Mann-Whitney U, and Wilcoxon signed rank test with paired data analyses were conducted using the statistical software R v 3.4.4 (R Core Development Team 2018).

Results

Trapping

P. clarkii were found at higher rates compared to *F. rusticus* in the southern site on the North Branch of the Chicago River (100% of 53 crayfish collected were *P. clarkii*). At the junction of the North Branch and North Shore Channel, *P. clarkii* consisted of 97.44% of the 39 crayfish caught, followed by 98.70% of 307 crayfish at the southern site of the Channel. *P. clarkii* were in lowest proportions in the northern most site on the North Shore Channel (18.67% of 332), compared to *F. rusticus*, and in Wilmette Harbor (6.25% of 32).

Across all individuals trapped *P. clarkii* were significantly larger (n = 360; average CL = 50.76mm) than *F. rusticus* (n = 157; 43.02mm) (t-test, p = <<0.001) (Figure 2). We observed *P. clarkii* to be more aggressive when captured but did not attempt to quantify this in the field. Males were captured at higher rates for both *P. clarkii* (65% of all captured) and *F. rusticus* (60%).

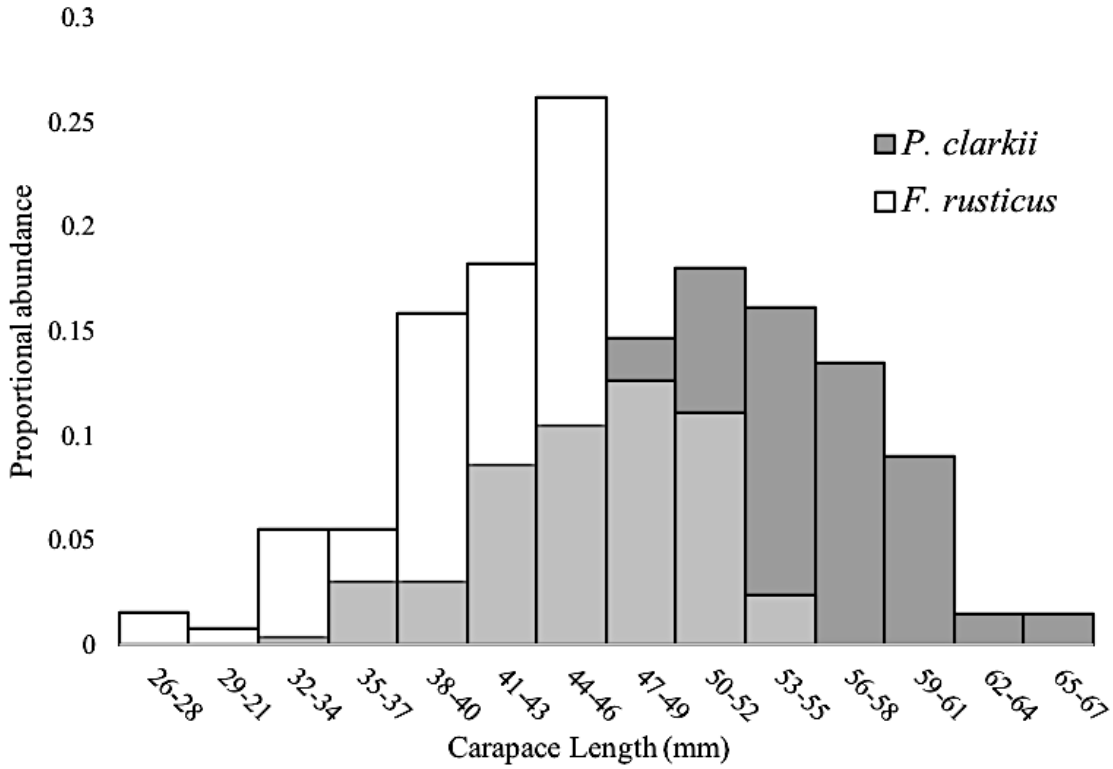


Figure 5. Distribution of carapace length of all crayfish in the North Shore Channel, North Branch, and Wilmette Harbor.

Competition Experiments

Shelter

Aggression was shown by raising the claws or grabbing the fish with claws. *P. clarkii* responded to fish attacks with aggression at least once during more trials (23/23) than *F. rusticus* (9/23) (X^2 with Yates correction, p = <<0.001). Fifty-six percent of *F. rusticus* that fled the fish attacks sought shelter during the trial. *F. rusticus* were significantly more likely to spend time in the shelter during the trial period (n=13/23) than *P. clarkii* (n=4/23) (X^2 with Yates correction, p = 0.015). However, once inside *P. clarkii* stayed in the shelter longer (t-test, $p = 0.017$). The four *P. clarkii* that entered the shelter spent an average of 16.01 minutes of the 20-minute trail inside. The 13 *F. rusticus* that entered the shelter spent an average of 7.67 minutes inside.

Interaction between *P. clarkii* and *F. rusticus* was recorded every 10 seconds during the entire trial (Table 1). *P. clarkii* displayed aggressive behaviors [1-5; Table 1] significantly more often than *F. rusticus* (Wilcoxon signed rank test with paired data, $p = 0.0424$). *F. rusticus* displayed submissive behaviors [-1, -2; Table 1] significantly more often than *P. clarkii* ($p = 0.0496$).

When competition was eliminated and trials with one crayfish per tank were conducted, *F. rusticus* entered the shelter forty-one percent of the trails compared to twenty-five percent of *P. clarkii.*

Table 1. Counts of behaviors shown by each species towards the other species during shelter trials. Behavior was recorded every 10 seconds throughout each 20 minute trial, but no count was made if crayfish were not interacting or nearby each other. Counts were summed among the 23 test animals of each species

Behavior	Level	Description	*Procambarus clarkii*	*Faxonius rusticus*
Submissive	-2	Fleeing, tail flip, walking away (rapidly)	2	0
	-1	Avoidance, walking away (slowly)	24	63
		Total submissive	***26***	***63***
Neutral	0	Within one body length of other crayfish but no interaction	135	154
Aggressive	1	Approach, turning toward	61	67
	2	Display, claws raised, antenna point	106	62
	3	Physical contact, claw touching, claw tapping (no grasping)	243	227
	4	Physical contact, claw grabbing	80	68
	5	Physical contact, claw ripping	6	2
		Total aggressive	***496***	***426***

Food

Although *P. clarkii* ate the food in more of the food competition trials (n=14/21) (X^2 with Yates correction, $p = 0.064$) and did so more quickly (average time to consumption of 58.21 minutes vs. 81.40 minutes for *F. rusticus*; Mann-Whitney U, $W = 35$, $p = 0.322$), neither of these measures were significantly different for the species. In all trials examining food competition the two crayfish showed aggressive behavior toward each other, indicating that competition was taking place**.**

Predation Experiment

We report predation rates as the percent of 24 hour trials after which we found crayfish had been removed from the tethers. *P. clarkii* were predated at a significantly higher rate (32%) than *F. rusticus* in (17%; X^2 with Yates correction, $p = 0.0496$) when both habitats were combined (Figure 3). This trend was also significant in the North Shore Channel, where 29% of *P. clarkii* were predated vs. 8% of *F. rusticus* (X^2 with Yates correction, $p = 0.0299$). Differences in predation rate were not significant when only Wilmette Harbor was considered (35% of *P. clarkii* predated vs. 24% *F. rusticus*; X^2 with Yates correction, $p = 0.360$). No difference was seen between the two sites for *F. rusticus* or *P. clarkii* (X^2 with Yates correction, *F. rusticus:* $p = 0.1047$; *P. clarkii:* $p = 0.567$). Turbidity levels in Wilmette Harbor were lower (average 206.5 cm secchi depth) than in the North Shore Channel (79.74 cm secchi depth).

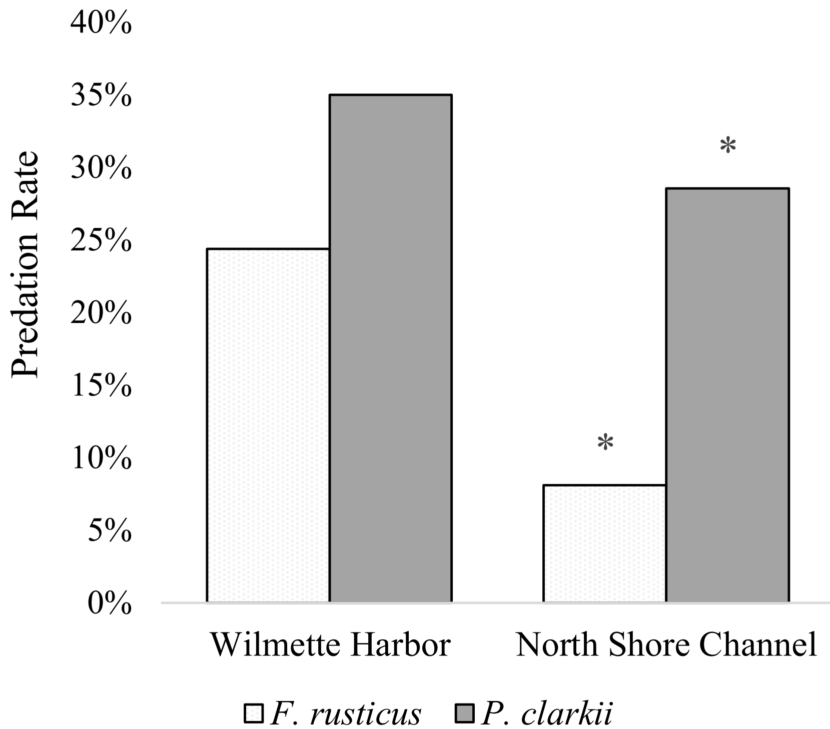


Figure 6. Predation rates on *F. rusticus* and *P. clarkii* in the North Shore Channel and Wilmette Harbor.

Discussion

P. clarkii is an internationally well-known invader of freshwaters, with populations established in Africa, Asia, Europe, and elsewhere in North America (Yue et al. 2010; Smith et al. 2018). It has been shown to be competitively dominant when it enters new systems (Yue et al. 2010) and has been associated with loss of native crayfish in several global regions. There is much concern about the spread of this species into the Great Lakes region, where it is most likely to interact with *F. rusticus* if it spreads. We have shown that *P. clarkii* is larger than *F. rusticus* and is more likely to behave aggressively when threatened. Although the food competition results were not significant, we note that *P. clarkii* ate the food in 2/3 of all trials when individuals were size-matched. In field situation the *P. clarkii* are likely to have a size advantage, meaning that they could potentially have an advantage in direct competition for food. Our shelter experiments showed that *P. clarkii* reacted aggressively to a simulated fish attack while *F. rusticus* was much more likely to flee and seek shelter. These results suggest that, in the absence of predators, *P. clarkii* will outcompete *F. rusticus*. Predation, however, appears to work in the opposite direction with *P. clarkii* being eaten at a higher rate than *F. rusticus.* This effect may restrict the spread of *P. clarkii* and may allow for coexistence of populations of these species.

Body size is a determinant of dominance in crayfish (Pavey and Fielder 1996; Issa et al. 1999) with larger individuals most often winning competitive interactions for resources (Butler and Stein 1985; Mazlum 2007). Trapping is size selective toward larger crayfish (Hein et al. 2007), meaning that our sampling is likely biased towards the largest individuals. We found that *F. rusticus* in the Channel and Wilmette Harbor are significantly smaller than *P. clarkii.* Their smaller size could result in *F. rusticus* being out competed by *P. clarkii* for any limiting resource.

We followed our field observations of crayfish size with lab experiments to measure competition between these species for two resources that have been found by other studies to be important predictors of dominance and survival (Figler et al. 1999; Gherardi and Daniels 2004). It is important to note here that we size matched individuals for these experiments, and thus that we likely gave an advantage to the smaller species (*F. rusticus*) relative to what would be experienced in the field. First, we tested for dominance at consuming a single piece of food when individuals of each species were starved and placed together. Although *P. clarkii* ate the food in two thirds of the food competition trials the result was not significant. In the field, *P. clarkii* are likely to display a size advantage, which has been found to be a determinate of fitness. This implies that although there was no difference between species during size matched food competition, there may be a significant difference in the field.

Next, we tested for dominance at accessing a single shelter when individuals of each species were threatened. In this experiment we found that *F. rusticus* entered the available shelter significantly more often than *P. clarkii*. While this is often concluded to be evidence of dominance because individuals will fight for the shelter, we observed few interactions between the species that suggested competition for this resource. Instead, when threatened by the artificial fish *P. clarkii* were significantly more likely to react with aggression towards the fish rather than flee, while the *F. rusticus* was more likely to seek shelter. We also observed *P. clarkii* to show aggressive behavior significantly more often in its interactions with the other crayfish. Our results also show that once inside a shelter *P. clarkii* stayed there for a significantly longer time. We do not have a good explanation for why this is, but suggest that it may be an underlying behavioral trait of unknown importance to competition. *P. clarkii,* unlike *F. rusticus,* are a burrowing crayfish species (Correia and Ferreira 1995), and remaining in the shelter during our experiments may have been akin to remaining in a burrow once one is found.

Our field observations of crayfish size combined with our competition experiments to indicate that *P. clarkii* is likely able to outcompete *F. rusticus* for limited resources. While our food experiment results were not statistically significant, we consider it likely that in the field – where crayfish are not size-matched – *P. clarkii* would win more competitions for food. This competitive ability is driven at least in part by the additional aggression shown by *P. clarkii* relative to *F. rusticus*. This interaction could be tested in lab using non-sized matched and mixed species trials. This aggression was significantly greater both in response to *F. rusticus*, and in response to our simulated fish attack.

Based on our sampling of population densities (Figure 1) and the competition results we hypothesized that the higher levels of aggression shown by *P. clarkii* may come at a cost in terms of higher predation. Specifically, responding aggressively may be useful when interacting with another crayfish, especially one from a species that is generally smaller. In contrast, this behavior may be detrimental when the threat is a much larger predator, such as one of the several species of fish that eat crayfish. Our sampling results showed that although *P. clarkii* are found throughout the North Shore Channel they are rarely found in Wilmette Harbor of Lake Michigan. These habitats are separated by a weir, but given the propensity of *P. clarkii* to travel overland (Ramalho and Anastácio 2015; Smith et al. 2018) we doubt that this is a serious barrier to movement, particularly given that the weir is occasionally opened to allow water to flow from the Channel into the Harbor.

We designed our predation experiment to test for differential levels of predation on the two crayfish species within and between the Channel and Harbor habitats. We hypothesized that predation on *P. clarkii* would be higher than on *F. rusticus* in both habitats, and that each species would face higher predation in the clearer water of the Harbor where visual predators, such as fish, should be more effective. Much of this hypothesis is supported by our results. Specifically, *P. clarkii* are significantly more likely to be predated across both habitats, and this was also true in just the Channel habitat. Both species were predated more often in the Harbor than the Channel, but this trend was not significant. Crayfish in this experiment were tethered to lines 30cm long. Although this would have limited their ability to flee from a threat, the experiments were conducted during late summer when macrophyte cover was plentiful in both habitats. Thus, we conclude it is likely that predation rates are somewhat inflated over what would occur without tethering, but that the difference between the species is indicative of true susceptibility to predation.

Evidence suggests that *F. rusticus* was established in the North Shore Channel before the invasion of *P. clarkii*. The latter is now dominant throughout the Channel but rarely found in the connected habitat of Lake Michigan. Although not fully conclusive, our results suggest that in the Channel *P. clarkii* dominance is driven by this species outcompeting *F. rusticus*, but that this competitive advantage is negated in the Harbor where predation on *P. clarkii* prevents them establishing a population. An alternative explanation for the distribution patterns observed is that the population of *P. clarkii* is still spreading within the Channel and will eventually move into the Harbor and Lake Michigan. Data are not available to test this but we consider it unlikely because of the high population of *P. clarkii* in much of the Channel which extends right up to the weir between the Channel and Harbor (Figure 1).

If there is a trade-off between competitive advantage and exposure to predation that explains the distribution patterns observed then we would expect *P. clarkii* to continue their spread into habitats that are either quite turbid, or that are clear but have few crayfish predators. This would make most rivers and wetlands across the Great Lakes region susceptible. While much of the Great Lakes themselves may be too clear, there are large areas that have secchi depths similar to those observed in the Channel. These include Lake Michigan's Green Bay (Qualls et al. 2013) and Lake Huron's Saginaw Bay (GLEC 2006). The only known populations of *P. clarkii* that have existed in the Great Lakes region over a long period are in wetlands connected to Sandusky Bay in the western basin of Lake Erie. *P. clarkii* were first recorded at the Winous Bay Shooting Club there in 1967, and the Resthaven Wildlife Area in 1982 (Nagy et al. 2018). Both populations persist in the wetlands, and in neither case is there evidence that *P. clarkii* have moved into the clearer water of Lake Erie.

P. clarkii is a widespread invader internationally that has been associated with large negative impacts. Populations have become established in North America, Africa, Asia, and Europe (Donato et al. 2018; Smith et al. 2018). The invasion of *P. clarkii* in Africa has resulted in the reduction of macrophyte species and damaged shorelines (Smart et al. 2002). In Asia, the species' burrows have led to damaged irrigation systems resulting in poor crop yields, causing economic loss (Yue et al. 2010). *P. clarkii* can be a vector for disease and spread the crayfish plague which is lethal to crayfishes and has resulted in biodiversity loss in Europe (Donato et al. 2018).

P. clarkii is a recent and spreading crayfish invader of freshwaters in the Laurentian Great Lakes region (Nagy et al. 2018). There is much concern about its potential impacts and a desire to prevent its further spread. If it does continue to spread it will come into contact and competition with established invasive crayfishes, and most often this will be the widely established and currently dominant *F. rusticus*. Our work shows that *P. clarkii* are larger and more aggressive than *F. rusticus*, and that when threatened they are less likely to seek shelter. A consequence of this aggression, however, is that *P. clarkii* respond to threats – such as predators – by aggressively displaying their chela rather than fleeing. Our experiments and observations offer a mechanistic explanation for patterns in distribution of *P. clarkii*, and can be used to aid predictions of future spread.

CHAPTER III

CURRENT STATUS AND DISTRIBUTION OF NATIVE AND NON-NATIVE CRAYFISHES IN THE CHICAGO REGION

Introduction

Crayfish often act as ecosystem engineers in freshwaters (Glon et al., 2017). Because of their large effects, non-native species of crayfish pose large threats to ecosystem function and native biodiversity (Strayer & Dudgeon, 2010). At least 28 species of crayfish are considered non-native worldwide, with multiple considered to be invasive (Gherardi, 2010; Lodge et al. 2012). Despite this, in the Laurentian Great Lakes region the distributions and ecological roles of native and non-native crayfishes have been poorly studied (Peters et al., 2014). There is reason to believe that crayfishes have large impacts in the Great Lakes, however, because multiple native species are expanding their ranges, and two non-native species where introduced and established (Peters et al., 2014).

Around the world invasive crayfishes have large ecological and economic impacts, including damaged fish populations (Quinn and Janssen, 1989; Janssen et al., 2005), reduction in diversity and density of macrophyte communities (Lodge et al., 1994; Wilson et al., 2004), decreased water quality (Albertson and Daniels, 2016; Souty-Grosset et al., 2016) and altered macroinvertebrate communities (Wilson et al., 2004; Albertson and Daniels, 2016). Invasive crayfishes compete with native species for food and shelter, often displacing native crayfish species (Quinn and Janssen, 1989; Janssen et al., 2005). In the Great Lakes, crayfish predate lake trout and lake sturgeon eggs, decreasing their population size and potentially resulting in damaged commercial and recreational fishing opportunities (Claramunt et al., 2005; Jonas et al., 2005; Fitzsimons et al., 2007). The rusty crayfish (*Faxonius rusticus*), an invader in parts of the Great Lakes region, can alter sediment accumulation in streams and alter habitat for benthic organisms, and increase water turbidity (Albertson and Daniels, 2016).

Thirteen species of crayfish have been recorded in the Great Lakes region (Taylor et al., 2015; Peters et al., 2014). Two of these – the obscure crayfish (*Faxonius obscurus*) and the red swamp crayfish (*Procambarus clarkii*) – are non-native to the whole Great Lakes. *F. obscurus* is native to the midwestern United States and has spread to the eastern United States and Ontario, Canada. *P. clarkii* is native to the southern United States, with a native range reaching the southern tip of Illinois. An additional three species are native to some part of the Great Lakes but have spreading populations (Peters et al. 2014). The rusty crayfish (*F. rusticus*) is considered native in western Ohio, eastern Indiana, and northern Kentucky. This species has spread – primarily through use as a bait organism – and it is now invasive throughout much of the Great Lakes basin (Peters et al., 2014). Calico Crayfish (*Faxonius immunis*) and Northern Clearwater Crayfish (*Faxonius propinquus*) are native to the Great Lakes Region and their range is also believed to be expanding (Peters et al., 2014).

The present study is focused on Lake Michigan where seven species of crayfish have been recorded (Peters et al., 2014). Two of these are non-native to the whole lake – *F. rusticus* and *P. clarkii*. The five native species are the devil crayfish (*Lacunicambarus diogenes), F. immunis, F. propinquus,* virile crayfish (*Faxonius virilis),* and the White River Crayfish (*Procambarus acutus*). *F. rusticus* is widely distributed in Lake Michigan and its catchment and is often found at high densities (Peters et al. 2014). *P. clarkii* is a recent invader of the southern catchment of the lake where its arrival results in new interactions between itself, native crayfishes, and *F. rusticus*.

Despite the negative impacts that invasive crayfish can have, the distribution of crayfishes in the Great Lakes in general, and in Lake Michigan, is not well resolved (Peters et al., 2014). In Lake Michigan and its tributaries as a whole, Peters et al. (2014) could find just 423 records of crayfish sampling over the 126 years between 1882-2008. The present study is focused on the southern basin of Lake Michigan (Figure 7) from where Peters et al. (2014) reported just 38 records, the first in 1906 (Peters et al., 2014; Figure 8; Appendix Table 2). Some of these sites have been sampled multiple times, but the long period covered and low number of samples clearly shows that crayfish sampling has been historically limited in this region.

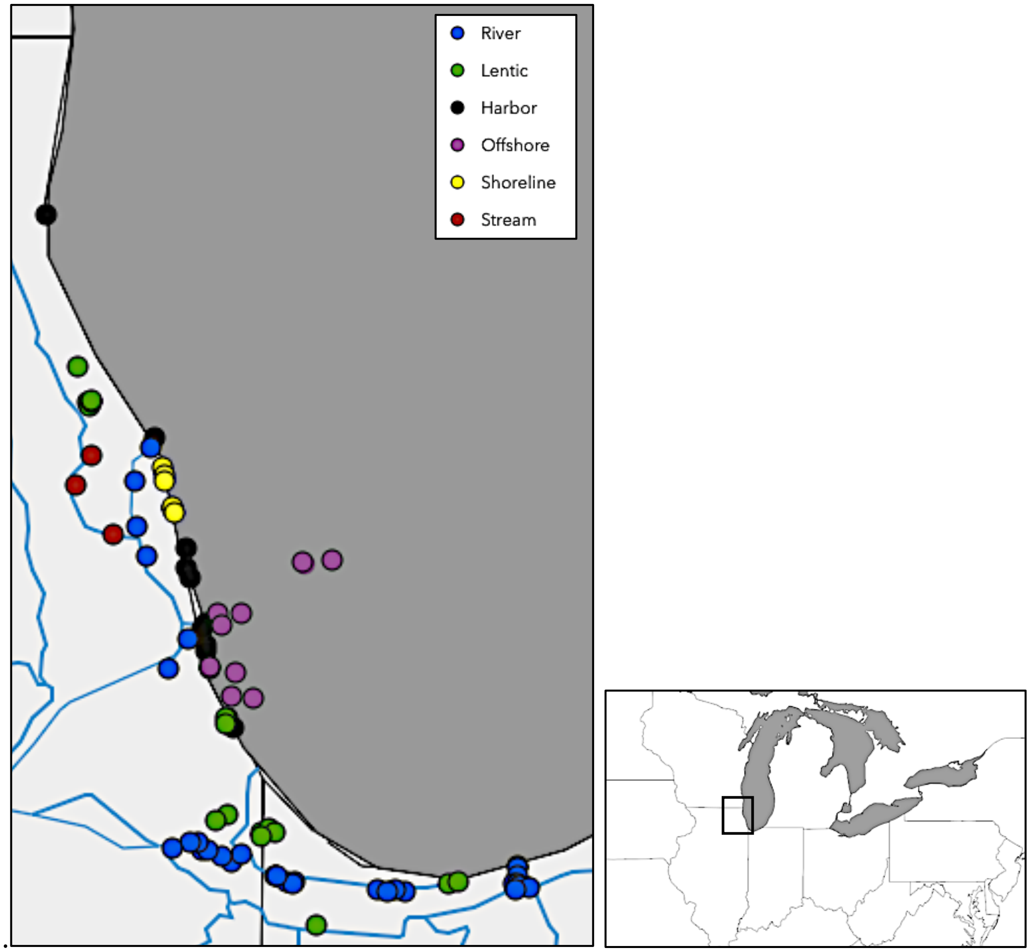


Figure 7. Sites sampled for crayfish in the Chicago region in Illinois and Indiana between 2015-2018, colored by habitat type (stream, river, lentic, shoreline, harbor, and offshore in Lake Michigan).

Figure 8. Sites sampled for crayfish in the Chicago region in Illinois and Indiana, as reported in Peters et al., 2014, during the years of a) 1906-1979, b) 1980-1989, c) 1990-1999, d) 2000-2008.

The southern basin of Lake Michigan and the Chicago region is of particular importance for invasive species study due to the large range of freshwater habitats, the large human population, and the effects of climate change. Habitats in the region include small streams, large rivers, inland lakes, harbors, and deep areas of Lake Michigan, each providing different opportunities for crayfishes to become established. The large human population of the Chicago Region results in a high introduction pressure (Capinha et al., 2013) from multiple vectors through which non-native freshwater species can be introduced by humans. These vectors include the pet trade, releases of classroom pets, boating and fishing, aquaculture, and live food (Keller & Lodge, 2007). Finally, the effects of climate change will create new thermal habitat in the Chicago region. Due to its location at the southern tip of the Great Lakes Region, this area will likely see the highest water temperatures of Lake Michigan (GLISA 2014). This change in thermal habitat could allow for the establishment of new non-native species, including crayfishes (Bellard et al. 2013).

Within species there is likely diversity in habitat selection. Larger crayfish tend to inhabit deeper waters, whereas smaller crayfish change their depth preference based on predator abundance (Englund & Krupa, 2000; Clark et al., 2013). Additionally, juvenile crayfish may rely more heavily on habitat that provides shelter to avoid predation from fish, and the presence of large rocks has been shown to significantly increase survival of small crayfish (Clark et al., 2013). In combination with the wide range of freshwater ecosystems across our study region, this presents the potential for a wide range of associations between crayfishes and habitats.

In this study we sampled crayfish in a range of freshwater habitats including offshore in Lake Michigan, rivers in Illinois and Indiana, inland lentic systems, streams, harbors, and the shoreline of Lake Michigan (Figure 7). We aimed to determine the species of crayfish found across this region and to examine the role of habitat type in influencing the distribution of native and non-native species. Our study area ranged from Waukegan in the state of Illinois to the

Indiana Dunes National Lakeshore in Indiana, thus spanning approximately 140 km of the southern basin of Lake Michigan.

Methods

Habitat Types and Sampling Effort

The Chicago region contains a diverse range of freshwater ecosystems which we delineated into six different habitat types. Three of these were inland (i.e., not in Lake Michigan): streams (<1m depth, generally fast flowing), rivers (>2m depth, flowing but not rapidly except during floods), and lentic (lakes). Within Lake Michigan three additional habitats were sampled; shoreline (along the shore, not in a harbor), harbors (enclosed boat harbors with direct access to Lake Michigan), and offshore (any sites not directly on a shoreline or in a harbor).

Seventy-eight sites were sampled for crayfish over the course of four summers (2015 – 2018), with sampling effort roughly proportional to the availability of each habitat type (Figure 7). In total we sampled at three stream, 31 river, 19 lentic, six shoreline, 12 harbor, and nine offshore sites. All stream sites were in Cook County Forest Preserves on the North Branch of the Chicago River. River and lentic sites were sampled throughout the Chicago region. We sampled in all harbors to which we could gain access. Shoreline sites were generally on the north side of Chicago and included a range of habitat types (concrete, steel and rock walls, piers). Offshore sites ranged from 1 - 13 km from shore, corresponding to a range of depths from 3.3 - 23 m. All offshore sites were in areas that we believed to have appropriate habitat for crayfishes, including rock breakwalls which extended into Lake Michigan, boat wrecks, and rocky reefs.

A total of 2,681 traps were set during 109 sampling events, where each event was a single site on a single date. Sampling occurred from May-October in 2015 (N=18 sites), 2016 (N=22), 2017 (N=26), and 2018 (N=51). Nine sites were sampled during two years, five were sampled during three years, and six were sampled during all four years.

Sampling Methods

In stream, river, lentic, shoreline and harbor sites, 15-20 standard minnow traps with openings enlarged to ~3.5 cm (Hein, 2006) were placed ~10 m apart and attached to either floats or objects such as trees or posts. In harbors, traps were tied off to docks or floats and placed throughout the harbor. Traps were set by wading, from a boat, or from docks, depending on habitat.

The depth at which traps were set depended on habitat type. In streams the traps were generally in 0.5 m of water, and never more than 1m. In rivers traps were placed at depths from 0.5-3.8 m, and in lentic systems from 0.3-2.6 m. All shoreline sites were in 0.5 m of water, except for traps placed around the Loyola Pier site which were at depths of up to 1.9 m. All minnow traps were baited with ~100 g dry dog food and left overnight before being recovered the next day.

In 2016, two offshore sites were sampled with 20 minnow traps each, but we did not catch any crayfish using this method. These sites were sampled by SCUBA divers laying traps baited with dog food, and retrieving the traps 24 hours later. We observed that round goby (*Neogobius melanostomus*) immediately entered the traps in large numbers and we suspect that they consumed all the bait before crayfish could enter. In subsequent summers (2017, 2018) we sampled by hand, capturing crayfish under rocks that we turned over and searching for them among debris of boat wrecks.

For all crayfish captured at all sites we determined species, sex, and size. Size was measured as carapace length (distance from the tip of rostrum to end of carapace) using manual calipers. The *Field Guide to Crayfishes of the Midwest* (Taylor et al., 2015) was used for species identification. Crayfish were released after data was taken.

Catch Per Unit Effort

In sites sampled by minnow trap we calculated catch per unit effort (CPUE) by determining the average number of crayfish caught per trap. CPUE for offshore sites sampled by hand catching was measured as the number of crayfish caught per minute of effort. Minutes of effort was calculated by adding the amount of time spent searching by each diver at each site. CPUE for offshore sites is not directly comparable to CPUE for other sites.

Data analysis

ANOVA and Tukey's HSD analyses were conducted using the statistical software R v 3.4.4 (R Core Development Team 2018). Maps were created using ESRI ArcGIS v 10.6.1 (ESRI 2018).

Results

Crayfish were found in all six habitat types. A total of 1,753 individual crayfish from six species were captured during this study (Table 2). *F. rusticus* and *P. clarkii* are both invasive to the region and were found in highest numbers. The remaining four species are native.36 sampling events resulted in no crayfish being caught. These sites varied by habitat type and tended to be some of the most highly disturbed. For example, no crayfish were caught in Bubbly Creek, a branch of the Chicago River with a long history of industrial pollution. We did not find trends in crayfish density or communities among years at any sites and pooled all data from each individual site for analysis (see Appendix 1, Figure 7 & Table 2, for full data).

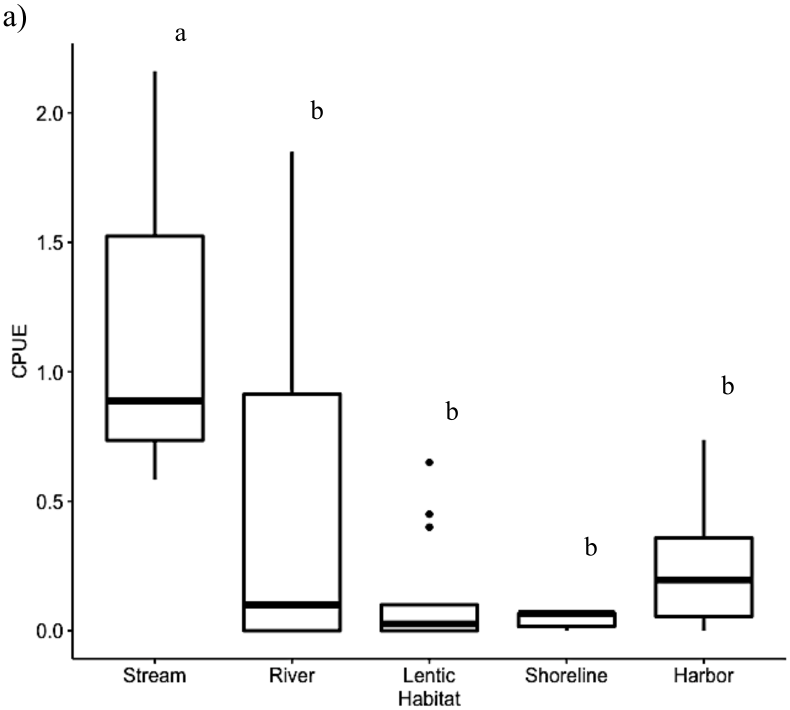
There was no significant variation in the number of crayfish caught based on the time of year that we sampled (Figure A1). Relatively low numbers were caught during our few sampling events in the month of May, but these were no lower than many samples taken in other months. We were able to catch high densities of crayfish as late as we sampled, which was early October (see Appendix 1, Figure 7).

Table 2. Catch per unit effort (CPUE) for each crayfish species within each habitat type in the Chicago region. – indicates that the species was not found in that habitat.
*Note that CPUE for offshore sites in Lake Michigan is not directly comparable to CPUE for other habitats. See methods for details.

	Total # found	**Stream CPUE**	**River CPUE**	**Lentic CPUE**	**Shore-line CPUE**	**Harbor CPUE**	**Offshore* CPUE**
Faxonius rusticus	1,021	0.986	0.354	0.021	0.038	0.193	0.201
Procambarus clarkii	503	-	0.329	0.044	-	0.010	-
Faxonius virilis	141	0.007	0.065	0.031	-	0.061	0.001
Faxonius propinquus	25	-	-	-	-	-	0.021
Faxonius immunis	42	0.296	-	-	-	-	-
Procambarus acutus	9	0.049	0.001	-	-	-	-

There was a significant difference in overall (i.e., all species combined) CPUE among habitat types excluding offshore sites (Figure 9a; ANOVA, $F_{(4,64)} = 4.71$, $p = 0.025$), with stream sites having significantly higher CPUE than harbor, lentic, and shoreline sites (Tukey’s HSD, p = 0.0144, 0.0043, 0.007 respectively). This pattern was driven by the most commonly found crayfish – *F. rusticus*, with a similar pattern of significance when comparing CPUE of just this species across habitats (Figure 9b; i.e., ANOVA, $F_{(4,64)} = 4.56$, $p = 0.0027$). Stream sites had significantly higher (Tukey’s HSD) CPUE than harbor ($p = 0.0141$), lentic ($p = 0.0010$), river ($p = 0.0106$), and shoreline ($p = 0.0060$) sites; Figure 9b). When *F. rusticus* was excluded there was no significant difference in CPUE among habitats (ANOVA, $p = 0.412$).

a)



b)

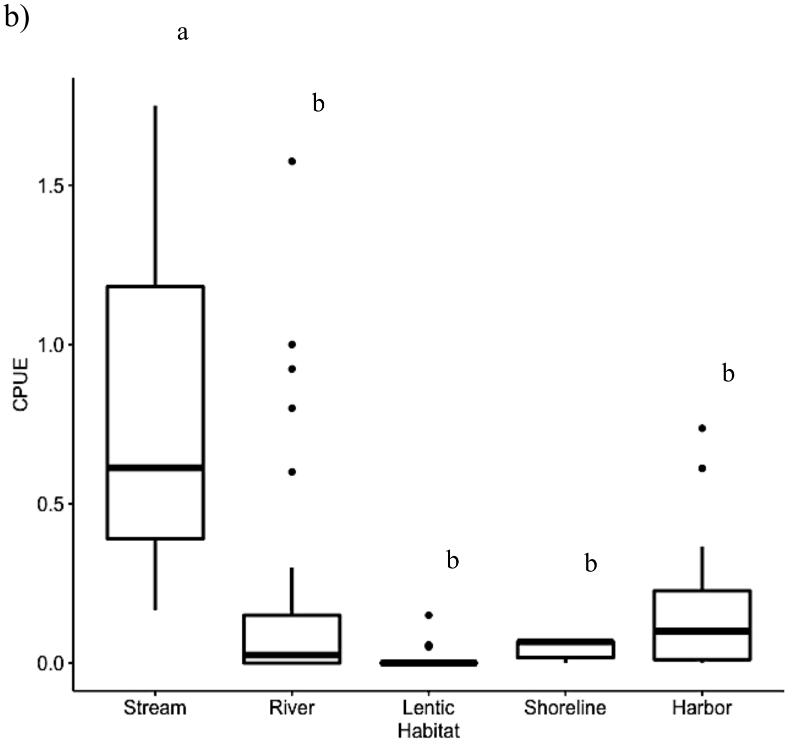


Figure 9. a) Density of crayfish (average #/trap) caught in each habitat type surrounding the Chicago region in Illinois and Indiana, b) Density of *F. rusticus* crayfish (average #/trap) caught in each habitat type.

Species Distributions

All habitat types were dominated by invaders (Table 2; Figure 10). *F. rusticus* was the most common and widely distributed species and was found at highest CPUE in all habitats except lentic (Figure 10; Figure 11a). Across all stream and harbor sites *F. rusticus* was found at least three times as often as any other species. *P. clarkii* were primarily found in river sites, including the North Shore Channel and North Branch of the Chicago River (Figure 11b). This artificial canal runs from Wilmette Harbor north of Chicago and connects to the North Branch of the Chicago River. *P. clarkii* was also found in small numbers (n=2) in Wilmette Harbor which is connected to the North Shore Channel, and in Jackson Park Harbor which connects to Lake Michigan on the south side of Chicago (Figure 11b).

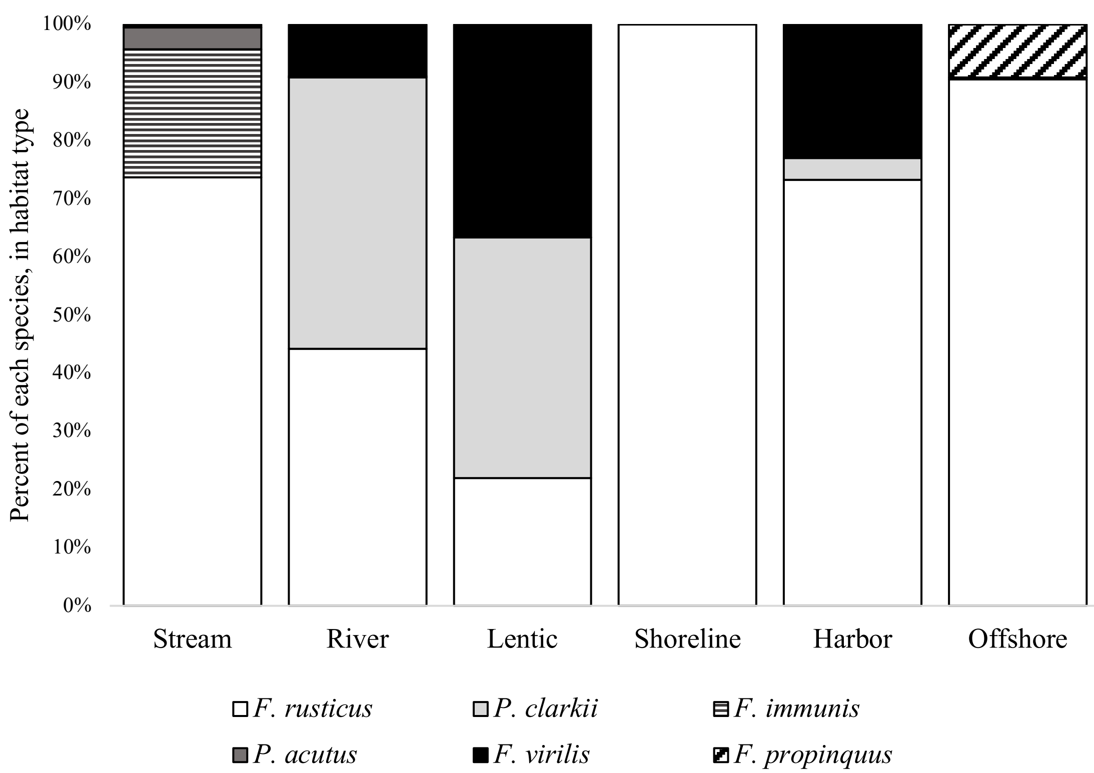


Figure 10. Percent of each crayfish species in each habitat type found in the Chicago region, in Illinois and Indiana

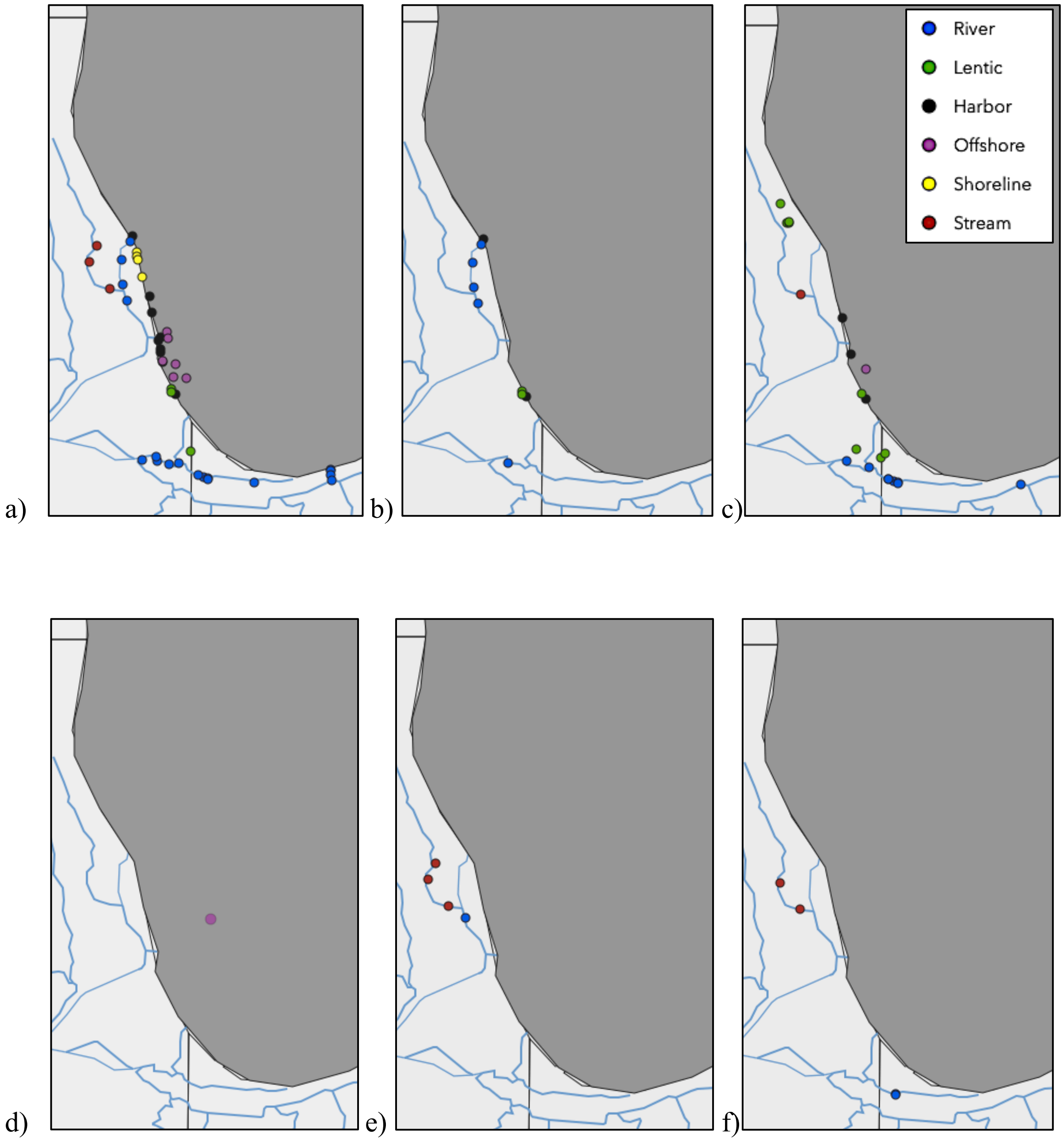


Figure 11. Distribution of a) *Faxonius rusticus,* b) *Procambarus clarkii,* c) *Faxonius virilis,* d) *Faxonius propinquus,* e) *Faxonius immunis,* f) *Procambarus acutus* in the Chicago region, colored by habitat type.

Four native species were found during this study. *F. virilis* was the most common of these and was found at 25 sites (Figure 11c). This species was found in streams, rivers, lentic sites, harbors, and one individual was found at an offshore site (Figure 11c). The only non-offshore sites where native species were numerically dominant were the lentic Chicago Botanic Gardens and three locations within the lentic Skokie Lagoons where *F. virilis* was the only species found. *F. virilis* was found in low densities at these sites, with CPUEs of 0.0571 (Botanic Gardens), and 0.071, 0.050, 0.40 (Skokie Lagoons sites 1, 2, and 3). At one additional Skokie Lagoons site we found no crayfish. Sites within Skokie Lagoons and the Botanic Gardens had deep mucky substrate and high density of macrophytes.

The other native species found were *F. immunis* (Figure 11e) and *P. acutus* (Figure 11f). *F. immunis* crayfish were found at three stream sites and one river site, and *P. acutus* were only found at stream sites. We note, however, that the seven *F. immunis* at the river site were found during sampling in 2018 that was being conducted for a different study in the Keller lab. *P. acutus* were found at low densities in two additional sites on the Grand Calumet River in Indiana (each with a CPUE of 0.050).

Streams within the Cook County Forest Preserves were sampled during all four years. These sites contained the highest diversity of crayfishes and contained more native species than other habitat types. *F. rusticus* were also found at high densities in stream sites (Table 2). *F. immunis* were commonly found in streams but at roughly one third the CPUE of *F. rusticus* (Table 2). *F. virilis* and *P. acutus* were found at low densities in these streams (Table 2).

Species Distributions (offshore)

Nine offshore sites were sampled by SCUBA diving. A majority of crayfish found at offshore sites were *F. rusticus* (Figure 11a; Table 2). The only site sampled via SCUBA diving that was dominated by native crayfish was a limestone bed located 13 kilometers offshore at a depth of 16 meters (Figure 11d). Twenty-five *F. propinquus*, and no other crayfish, were hand caught by two divers over a 40-minute period. This was the only site at which this species was found during the study. One offshore site, a shipwreck located at 25 meters resulted in no crayfish caught. In the remaining sites sampled by SCUBA, all sites (except the site with only *F. propinquus*) contained *F. rusticus*. The CPUE of *F. rusticus* ranged from 0.011 to 0.416. A single *F. virilis* was found while searching for crayfish on a wreck at 9 m depth.

The offshore sites that contained a higher density of crayfishes were dominated by rocky substrate that allowed crayfish to hide. The crayfish were most often caught by overturning rocks to reveal a hiding crayfish, these individuals were then caught with nets or by hand. Individuals were occasionally found on sandy substrate, but only when this was in close proximity to hard substrate. The highest CPUEs of crayfish caught via SCUBA were found at shallow depths of 3.5-5.5 meters. The deepest site where crayfish were caught was 16 meters.

Size comparison of species

We found significant differences among species in the size of crayfish caught using minnow traps (ANOVA, $F_{(4,1380)}$=232.5, $p < 0.001$). *P. clarkii* were significantly larger than all other species (Figure 12; Tukeys HSD; $p = < 0.001$ when compared to *F. rusticus, F. immunis,* and *F. virilis;* $p = 0.002$ when compared with *P. acutus*). *P. acutus* were significantly larger than *F. immunis* (Figure 12; Tukeys HSD; $p = 0.017$). Both *F. rusticus* (Tukeys HSD; $p = 0.019$) and *F. virilis* (Tukeys HSD; $p = 0.001$) were also significantly larger than *F. immunis* (Figure 12).

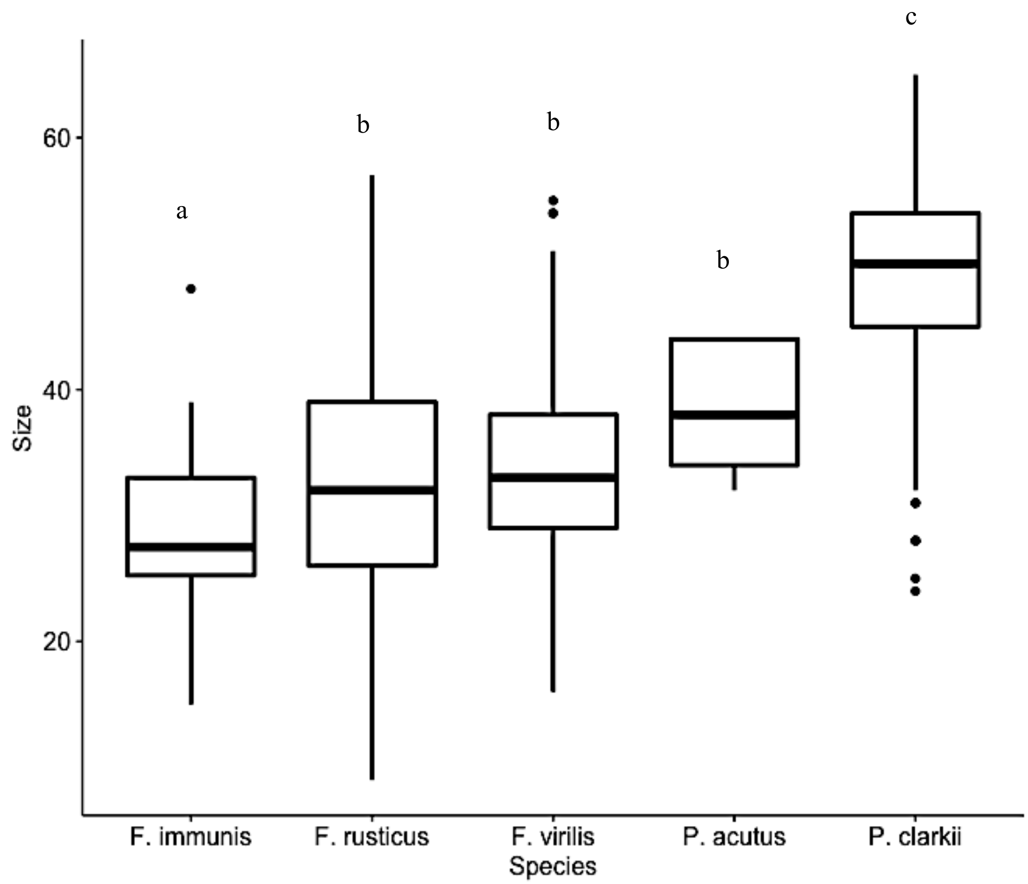


Figure 12. Size distribution of the carapace length (mm) of crayfish species caught in minnow traps in the Chicago region.

F. rusticus was the only species caught widely enough to compare its size across habitats. Comparing size among sites where it was captured by minnow trap and where it was hand caught is complicated because minnow traps are known to select for larger individuals from the population. To correct for this, we compared the size of *F. rusticus* captured in minnow traps to the largest 50% of *F. rusticus* caught by hand at offshore sites. This analysis showed that *F. rusticus* caught inland via minnow trap were significantly larger than the largest 50% of *F. rusticus* caught via hand catching in Lake Michigan (t-test; $p = << 0.001$). *F. rusticus* sizes were also significantly different between inland habitat types (all sampled by minnow trap; Figure 13; ANOVA, $F_{(5,841)}=78.27$, $p < 0.001$). Harbor *F. rusticus* were the largest, significantly larger than those found at lentic and stream sites (Figure 13; Tukeys HSD; p = <<0.001 for both habitat types). The second largest were those found in river sites, which were significantly larger than stream *F. rusticus* (Figure 13; Tukeys HSD; p = <<0.001).

F. virilis was found in fewer habitats, but those captured in harbors were significantly larger than *F. virilis* found in lentic (Tukeys HSD; p = 0.019) and river sites (Tukeys HSD p = 0.001).

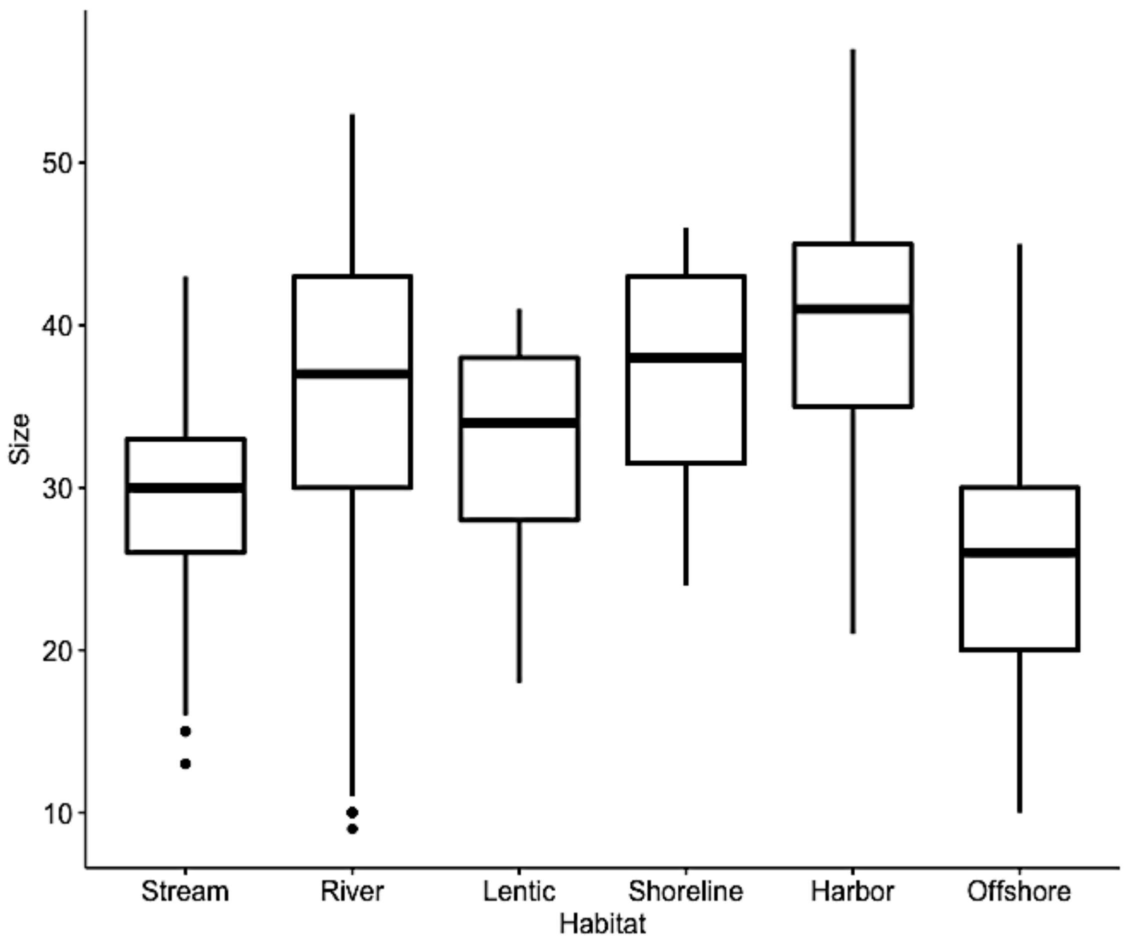


Figure 13. Size distribution of the carapace length (mm) of *Faxonius rusticus* caught in the Chicago region. *Lake Michigan crayfish were caught via hand catching; other habitat types were sampled via minnow trap.

Overlap in distributions

Three species found during this study were never found alone: *P. clarkii, F. immunis,* and *P. acutus*. *P. clarkii* most often overlapped with *F. rusticus*, and this occurred at nine sites (Appendix 1, Figure 8). At three sites *P. clarkii* co-existed with native species, including *F. virilis* (Jackson Park Harbor [harbor] and Jackson Park Lagoon [lentic]) and *F. immunis* (North Branch Chicago River in 2018). *F. rusticus*, which was found alone in 20 sites, was the only species commonly found alone. This species also overlapped with every other species in the study, with the exception of the *F. propinquus* which was only found at one offshore site. The stream sites within the Cook County Forest Preserves showed the highest species richness, with the maximum number of species found at a site being LaBagh Woods with four species.

Discussion

Despite their ecological and economic importance, the diversity and distribution of crayfishes is poorly known in the Great Lakes region (Peters et al., 2014). Peters et al. (2014) published a dataset of all known crayfish sampling data for the Great Lakes. This dataset showed that both invasive and native species were spreading, and that some native species declined. Although these general patterns are known, Peters et al. (2014) clearly demonstrated that there has been insufficient monitoring to determine either timing, rate, or impact of these changes. In this study we have more than doubled the number of sites sampled for crayfish in the Chicago Region and southern basin of Lake Michigan and have resampled many of these sites over multiple years.

Invasive crayfish distribution

Two invasive crayfish species were found. *F. rusticus* is considered as a range expanding species in the United States and is often termed invasive (Peters et al. 2014). This species has been found to reduce fine sediment accumulation, increasing water turbidity, reduce fish populations, damage macroinvertebrate and macrophyte communities, and decrease the abundance of native crayfishes (Wilson et al. 2004; Kreps et al. 2016; Albertson & Daniels 2016). *F. rusticus* are flexible in the habitats they live in and a total of 1,021 were captured during this study. They are the only species that was found in all habitats, and they were numerically dominant in all habitats except lentic. The first recorded occurrence of *F. rusticus* in our study area was in 1984 (Peters et al. 2014). In the 1990's and 2000's, the population appears to have spread because it was found at multiple sites along the shoreline (Peters et al. 2014). Our data show that *F. rusticus* are much more widespread in the region than previously reported. This may be due to actual spread in recent decades, insufficient sampling prior to our work, or a combination of both.

At 20 of our sampling sites *F. rusticus* was the only species collected. This species had the highest CPUE of all species in the study. Offshore Lake Michigan sites usually only contained *F. rusticus.* Crayfish were caught at a total of eight offshore sites, and six of these were dominated by *F. rusticus*. At one site, only three individuals were caught, two of which were *F. rusticus.* In all shoreline sites *F. rusticus* were the only species found. These sites were often characterized by rocky habitat without vegetation. Sites that had a sandy substrate and no shelter did not have any crayfish, presumably because the lack of shelter exposes crayfish to predation.

P. clarkii, a recent invader in the Chicago region, was found at nine sites and were the largest of all species found. This species is an international invader, causing decreased diversity of macrophytes, mollusks, and amphibians in many of the systems they have invaded (Souty-Grosset et al. 2016). *P. clarkii* increase water turbidity and decrease stream bank stability via burrowing, which can also damage water drainage systems and hinder restoration efforts (Anastácio and Marques, 1997; Rodríguez et al., 2003; Barbaresi et al. 2004). Additionally, this species is considered an agricultural pest because its burrows damage irrigation systems and individuals have been found to eat juvenile rice plants (Anastácio et al. 2005; Yue et al. 2010; Lodge et al. 2012). They have established small, localized populations in a southern harbor, lagoon, and river (Figure 11b).

The first record of this species in our study area is from 2001, in the Northern part of Illinois, near the border of Wisconsin and Illinois (Peters et al 2014). This was the only noted population of *P. clarkii* found in Peters et al. (2014). We found that this single population is not representative of the current spread of *P. clarkii.* Through our sampling, we found *P. clarkii* in two areas along the Lake Michigan shoreline, as well as at inland sites. We did not find evidence that these populations are actively spreading, but we note that our overall study may not have been long or intensive enough to find such spread if it is occurring. Our 2015 sampling was the first to document a reproducing population of *P. clarkii* in the North Shore Channel, a canal in the northern part of our study area which we designated as river habitat. This population overlaps with a previously established population of *F. rusticus*, and we believe that these are the first known overlapping populations of these two well-known and highly damaging invaders. Both *F. rusticus* and *P. clarkii* are found in high densities and our previous experiments (See Chapter 2) show that they are likely competing for resources.

The wide distribution of *F. rusticus* means that any further spread of *P. clarkii* will likely result in continued interactions between these species. Our previous work (Chapter 2) shows that these species can co-exist. Despite this, the relatively short duration of our sampling program makes it impossible to determine whether this co-existence will persist. In the North Shore

Channel the canal sections closest to Lake Michigan are dominated by *F. rusticus* and have very few *P. clarkii*, while further from Lake Michigan this pattern is reversed. There is no apparent habitat difference that would account for this change in species assemblage; the canal has extremely low flow, is consistent in depth and width, and has a uniformly soft and mostly mucky substrate. Given the invasion history of the region (Peters et al. 2014) we are confident that *F. rusticus* was present in this waterway prior to the arrival of *P. clarkii*. Thus, it is possible that the overlap in these species is part of a longer term expansion of *P. clarkii* which may eventually fully dominate the habitat.

Native crayfish distribution

Four species native to the area were found during the study. *F. propinquus* was found at a single site, which was an isolated reef habitat offshore in Lake Michigan. These individuals were among the smallest caught during the entire study period. This species is native to all of the Great Lakes, and its range has expanded to western Lake Superior and within lakes in Wisconsin (Hill & Lodge, 1999, Peters et al., 2014). This species was not found inland during our study period. Historically, this species was found in 1979, 1982-84, 1992, 1994-96, 1998, 2007, and 2008 along the shoreline of Lake Michigan within in our study area (Appendix 1; Table 2; Peters et al. 2014). We did sample in nearby areas, where this species was historically found, however no individuals were found along the shoreline.

F. virilis, the most common and widespread native species found, is also native to all five Great Lakes. In the 1970's, this species was widespread throughout the shoreline of our study area (Peters et al. 2014). Since the arrival of *F. rusticus* in 1984 (Peters et al. 2014)*, F. virilis* was has decreased in occurrence along the shoreline (Peters et al. 2014). This trend continues through the 1990's and 2000's where low numbers of *F. virilis* have been reported from our study area (Peters et al. 2014). We note that the Peters et al. (2014) dataset does not include inland sites.

F. virilis is similar in size to the invasive *F. rusticus*, and crayfish size is known to be a determinant of competitive outcomes among individuals and species (Pavey and Fielder 1996; Issa et al. 1999). Although this species has most commonly been found in rocky habitat around the Great Lakes, in this study individuals were often in harbors with primarily sandy habitat or in mucky habitat with turbid water and a high density of macrophytes. Although commonly found at the same sites as *F. rusticus*, we found *F. virilis* in lower numbers. *F. virilis* has been found to be predated on at higher rates when compared to *F. rusticus* in a Wisconsin lake (Hill & Lodge, 1999). We believe that *F. virilis* individuals are potentially occupying habitats with less hard substrate to reduce direct competition with *F. rusticus*.

P. acutus and *F. immunis* were each found at just a few sites during our study. Our findings were consistent with the low populations of these species found in Peters et al. (2014). From the 1970's – 2014, both species were rarely reported anywhere in the Illinois and Indiana portion of Lake Michigan (Peters et al. 2014). We found *P. acutus* in a single river site in the southern portion of our sampling area, and in two stream sites on the North Branch of the Chicago River where it runs through the Cook County Forest Preserve. The stream sites in this area contain higher amounts of rocky substrate compared to other inland sites sampled and the water is less turbid. These sites are also typically urban and experience high flows during rain events. In comparison, our river sites all have heavily regulated flows and rarely experience high flow rates.

Between 2015-2017, we only found *F. immunis* in the Forest Preserve stream habitat sites. In 2018, some individuals of this species were found south of the Forest Preserves, and this may have been associated with removal of a dam. This dam separated the upstream 'stream' sections of the North Branch from the lower 'river' sections, and was at the confluence with the North Shore Channel. The North Shore Channel, an artificial canal, contributes most of the water to the river sections of the North Branch of the Chicago River. We only found *F. immunis* in the 'river' portion after the dam was removed in July 2018.

Peters et al. (2014) reported that the Devil crayfish (*Lacunicambarus diogenes*), a native species, is present in our study area. Our use of minnow traps as a method for sampling excludes crayfish which are primarily burrowers, such as the Devil crayfish. Thus, our results do not shed light on the current distribution of this species.

We observed some signs that *F. rusticus* and *F. immunis* may be hybridizing within the Forest Preserve streams. Some captured individuals had distinctive patterns on the tail and carapace, consistent with *F. immunis*, as well as large "rust" spots on each side of their carapace, which is seen in *F. rusticus.* These individuals were counted as *F. rusticus* in our study due to the lack of a deep notch in the moveable part of the cheli, which is distinctive in *F. immunis.*

Future needs

Although our study has greatly increased knowledge of crayfish distributions in this area we have also identified additional locations that should be sampled. We found that the shoreline and shallow offshore sites dominated almost entirely *F. rusticus.* Most areas of the shoreline do not contain good habitat for crayfishes (i.e., no shelter), but when individuals were found, these individuals were always *F. rusticus.* Our SCUBA sampling indicates that there may be potential for deeper areas to provide refuge from *F. rusticus* for native species. This may be because *F. rusticus* have thus far not spread to these areas, or because they are not able to live at these greater depths. More sampling in these deeper areas would help to determine the cause of the patterns that we observed. In turn, this may be important information for understanding the extent and effects of crayfish predation on fish eggs. There are several lake trout (*Salvelinus namaycusch*) spawning reefs in the southern basin on Lake Michigan where egg predation by crayfishes may be particularly harmful to ongoing efforts to restore this species (Redman et al. 2017).

Although *F. rusticus* is the most common invader in the region, we found it to be so widely distributed that there are likely few options to reduce its spread and impacts. In contrast, *P. clarkii* appears to be at an early stage of invasion and there may be opportunities to prevent its spread both within and beyond our study region. The arrival of *P. clarkii* was most likely due to intentional transport for trade (DiStefano et al. 2016). This species is common in the pet trade, is used widely as a classroom pet, and can be purchased live for food (Keller & Lodge 2007; DiStefano et al. 2016; Chucholl & Wendler 2017). We have heard anecdotally this this species is included in science curricula within schools including the Chicago Public Schools and nearby suburbs. We suspect that release from one or more of these vectors has been responsible for the establishment of the species in the region.

Eradication efforts aimed at invasive crayfish are difficult, expensive, and rarely successful (Gherardi et al. 2011). Therefore, efforts to prevent the spread and further release of *P. clarkii* may be more beneficial. Mechanical efforts at removal often involve large amounts of trapping which requires considerable manpower (Gherardi et al. 2011). Biological control can be implemented using predatory fish introductions or introduction of a biocontrol agent that kill crayfish (Edgerton et al. 2002; Hein et al. 2006; Hein et al. 2007; Gherardi et al. 2011). Biocontrol agents, such as virus's (ex. white spot syndrome virus (WSSV)), parasites, or fungi (ex. *Aphanomyces astaci*) are high risk because they are not host specific, resulting in possible spread to non-target organisms (Edgerton et al. 2002; Gherardi et al. 2011). Chemicals can also be used to lower crayfish populations, but so far the only available chemicals are general pesticides that have negative effects on all flora and fauna in the waterbody (Gherardi et al. 2011). Physical methods have been used to prevent the spread of *P. clarkii.* Pond drainage and pond liners that prevent burrowing have been attempted but are not always successful due to *P. clarkii'*s ability to survive out of water and to burrow (Gherardi et al. 2011). Preventing introduction and range expansion may be a more effective tool for controlling invasive crayfish populations (Lodge et al. 2012; Loureiro et al. 2015). Early detection and rapid assessment can aid in prevention of spread (Loureiro et al. 2015). Increased regulation of the live food, pet, and classroom trade can also aid in preventing the introduction of non-native crayfish (DiStefano et al. 2016).

The areas with the greatest diversity of native crayfishes were streams in the Cook County Forest Preserves. Additionally, the Skokie Lagoons (lentic) and Chicago Botanic Gardens (lentic) contained a single native species and no invaders. These could be targets for conservation through efforts to prevent the arrival of *P. clarkii* (all sites) and *F. rusticus* (lentic sites). Stream sites in the region, such as the Cook County Forest Preserves sampled in this study, should also be priorities for more sampling. South of the Forest Preserves we found high densities of *P. clarkii.* Previously, a dam created a barrier preventing direct access for *P. clarkii* to access the Forest Preserves. This dam was removed in the summer of 2018. Although it does not appear that *P. clarkii* have moved north into the stream sites, one dead *P. clarkii* was found in the Forest Preserves in August 2018. No live individuals were seen or caught. Efforts to intensively trap and remove *P. clarkii* near the site of the previous dam may help to prevent or slow the spread of this invader into the stream sites.

We also recommend increasing sampling efforts in inland ponds and lakes that are easily accessible to the public. These ponds include those on golf courses, retention basins, and at neighborhood parks. In Milwaukee, Wisconsin, three ponds in an urban area were found to have established populations of *P. clarkii* (Wisconsin DNR). Attempts to eradicate these populations included intensive trapping with minnow traps, lining the ponds to prevent crayfish burrowing, and poisoning the ponds with chlorine bleach and an insecticide (Behm 2009). The poisoning methods were found to be lethal to crayfish in water, but at least some survived in burrows (Bunk 2014). Eventually, one pond was filled in, and the other two ponds are being monitored with trapping and additional removal efforts are being studied (Bunk 2014).

Previous attempts to reduce the size of the invasive *F. rusticus* population were successful in Sparkling Lake, Wisconsin (Hein et al., 2006; Hein et al., 2007). These studies used a combination of intensive trapping and increased predatory fish populations to successfully reduce *F. rusticus* population sizes (Hein et al., 2006; Hein et al., 2007). Methods to reduce *P. clarkii* populations should be investigated further to determine the most effective way to reduce the size, extent, and spread of established populations. Additionally, the identification of sites to which invasive crayfish species, such as *P. clarkii,* could spread should be prioritized. These sites include waterbodies connected by waterways, areas in which crayfish can conduct overland travel. Perhaps most important is to determine areas of high potential transportation by humans, which likely include neighborhood ponds, creeks, golf course ponds, and near boat launches. Prioritizing these sites for population reduction and spread prevention could reduce the spread of non-native crayfish species.

CHAPTER IV

DISTRIBUTION AND HABITAT ASSOCIATIONS OF NATIVE AND NON-NATIVE MACROPHYTES AND MOLLUSKS IN THE CHICAGO REGION

Introduction

Anthropogenically altered ecosystems often have less suitable habitat for native species and more habitat that is suitable for non-native species (Airoldi et al. 2015). The effects of this have been best studied in terrestrial ecosystems. For example, that cities and urban environments tend to be similar across the globe, leading to a common suite of non-native species (e.g., rats, pigeons) that are dominant in these ecosystems (Gaetner et al. 2016). Likewise, road edges tend to be good habitat for non-native weedy plants, which are then able to spread along the roads to reach new areas (Christen & Matlack 2008).

The effects of anthropogenic alterations to marine ecosystems have been less well studied. Despite this, it is known that anthropogenically altered habitats are more likely to contain non-native species compared to natural reefs and substrate (Bulleri & Airoldi 2005; Glasby et al. 2007; Gittenberger & C. van der Stelt 2011; Airoldi et al. 2015; Bieler et al. 2017). In marine ecosystems where the number native species is higher than the number of non-native species, artificial habitats such as pontoons were found to have greater non-native species compared to natural rocky reefs (Galsby et al. 2007). Shipwrecks, sunk both intentionally and accidently, in the Florida Keys have been found to have high numbers of non-native mollusk species attached to them (Bieler et al 2017).

Although freshwater ecosystems have higher biodiversity per surface area compared to marine systems (Dudgeon et al. 2005), the role that urbanization and anthropogenically hardened substrate plays in non-native mollusk and macrophyte populations have been relatively poorly studied. Despite this lack of study, it is thought that reduced native biodiversity and increased non-native species richness often result from anthropogenic alterations to freshwater ecosystems (Airoldi et al. 2015). These alterations include changes in flow rate and water level, introduction of new structures, and channelization. Perhaps the best described effects in freshwater ecosystems come from regulation of rivers, where flow is often regulated through the construction of dams that can transform a flowing river into a habitat more like a series of slow-flowing lakes. This affects water quality, depth, and flow-rate, as well as sedimentation rates (Bunn and Arthington 2002; Nilsson et al. 2005). Each of these alterations to habitat changes interactions between species and their environment, and potentially also between species. In turn, this leads to changes in biodiversity (Bunn and Arthington 2002). River Murray in Australia is regulated by several dams, which has altered the flow and changed the ecosystem in many ways (Walker et al. 1992). Organisms such as snails and bivalves were once common in the system, but their populations decreased in size with increased flow regulation (Walker et al. 1992). Fish populations have decreased due to loss of habitat and decreased flooding, causing reduced fish spawning (Walker et al. 1992).

Freshwater ecosystems in and around the city of Chicago have been extensively modified by humans, including the destruction of habitats that were once prevalent (e.g., small streams and wetlands) and the creation of habitats that did not previously exist (e.g., harbors) (Hill 2000). The region now consists of inland (i.e., not Lake Michigan) waterways and lakes, a long shoreline of Lake Michigan, and offshore habitats. Lotic systems in the area have been extensively deepened and widened to accept the additional water that flows to them due to the ‘hardening’ of terrestrial environments (Hill 2000). In many cases these systems have also been straightened to reduce flooding and to enhance navigation (Hill 2000). Inland lotic systems have also been highly altered, for example Lake Calumet is now much smaller than its original size, is regularly dredged to keep it open for shipping, and it has received extensive pollution from the surrounding industrialized area (Wilson and Weng 2010). Other lakes are entirely man-made, such as the Skokie Lagoons which were dug in the 1930’s as a public works program (Forest Preserves of Cook County 2019).

Human modifications of habitat have also been important in Lake Michigan. The most obvious of these are directly along the lakeshore, where harbors are prevalent, fill has been used to ‘reclaim’ land, and where breakwalls are used extensively to trap sand and create beaches (Chrzastowski 1999). Anthropogenic changes extend offshore where many artificial reefs have resulted from intentional and unintentional sinking of boats. The effects of these anthropogenic habitat changes in Lake Michigan on local native and non-native biodiversity are almost entirely unknown.

Although anthropogenically altered freshwater habitats in the Chicago region are prevalent, little is known about how these habitat changes have affected native and non-native species presence and diversity. Understanding the impacts of these habitats, as well as their arrangement across space, may be particularly important for management existing and new invasive species. In the work described here we have characterized aquatic habitats of the Chicago region and extensively sampled some mollusks and macrophytes across the range of habitats identified. These taxa are of particular interest because of the importance of recreational boating and commercial shipping in the region, and the central role that these vectors have played in spreading these taxa (Wilson et al. 2006; Johnson et al. 2009; Matthews et al. 2014). Our goals were to characterize habitats of the region and to determine the species communities present in each habitat type. This enabled us to assess ecological connectedness across habitats and to estimate the most likely routes by which invasive species may spread.

Methods

Habitat Categories

The Chicago region contains a variety of freshwater ecosystems. From our experience working on these systems and additional survey work during this project we classified inland (i.e., not in Lake Michigan) freshwater systems into three habitat categories: *streams* (<1m depth, generally fast flowing), *rivers* (>2m depth, flowing but not rapidly except during floods), and *lentic* (i.e., lakes). Along the Lake Michigan shoreline we classified a further two habitats: *harbors* and *shoreline* (i.e., all shoreline except harbors). Finally, we classified all habitat not directly on the shoreline as *offshore*. A total of six habitat categories were created.

We acknowledge that our *shoreline* category simplifies a diversity of shoreline types. To ensure that we sampled across the full range of these we created an ArcGIS layer of the shoreline of the Illinois portion of Lake Michigan using Google Earth (Google Earth v 7.3.2, 2018) and the United States Engineer Research and Development Center's (ERDC) imagery (USERDC). We determined that the *shoreline* habitat category consists of sandy beach, terrestrial vegetation to the water's edge, river mouth, harbor, pier, large boulders, artificial rock wall, and artificial vertical wall. The vertical wall category consisted of walls composed primarily of metal or concrete.

Sampling: Locations

Selection of *shoreline* sites for mollusk and macrophyte sampling (see below for sampling methods) was driven by the GIS analysis described above (see Figure 14 for maps of all sampling sites). Based on the map created we focused on sandy beaches, piers (not within harbors), and rocky substrates because these made up a total of 64.36% of the Illinois shoreline.

Harbors also fall on the shoreline but were assessed as a separate habitat category (see above) because they provide a very different habitat. Artificial vertical walls make up a further 34.6% of the Illinois portion of Lake Michigan's shoreline. These walls are constructed of concrete or metal and experience high wave activity. We visited many (N= >20) such sites and always found that the habitat at the base of the walls to be mobile sand. Due to the difficulty and safety issues of sampling at these sites and the simplicity of their aquatic habitat we focused our sampling efforts on other areas (but see *Results*).

Macrophyte Sampling Locations Mollusk sampling locations

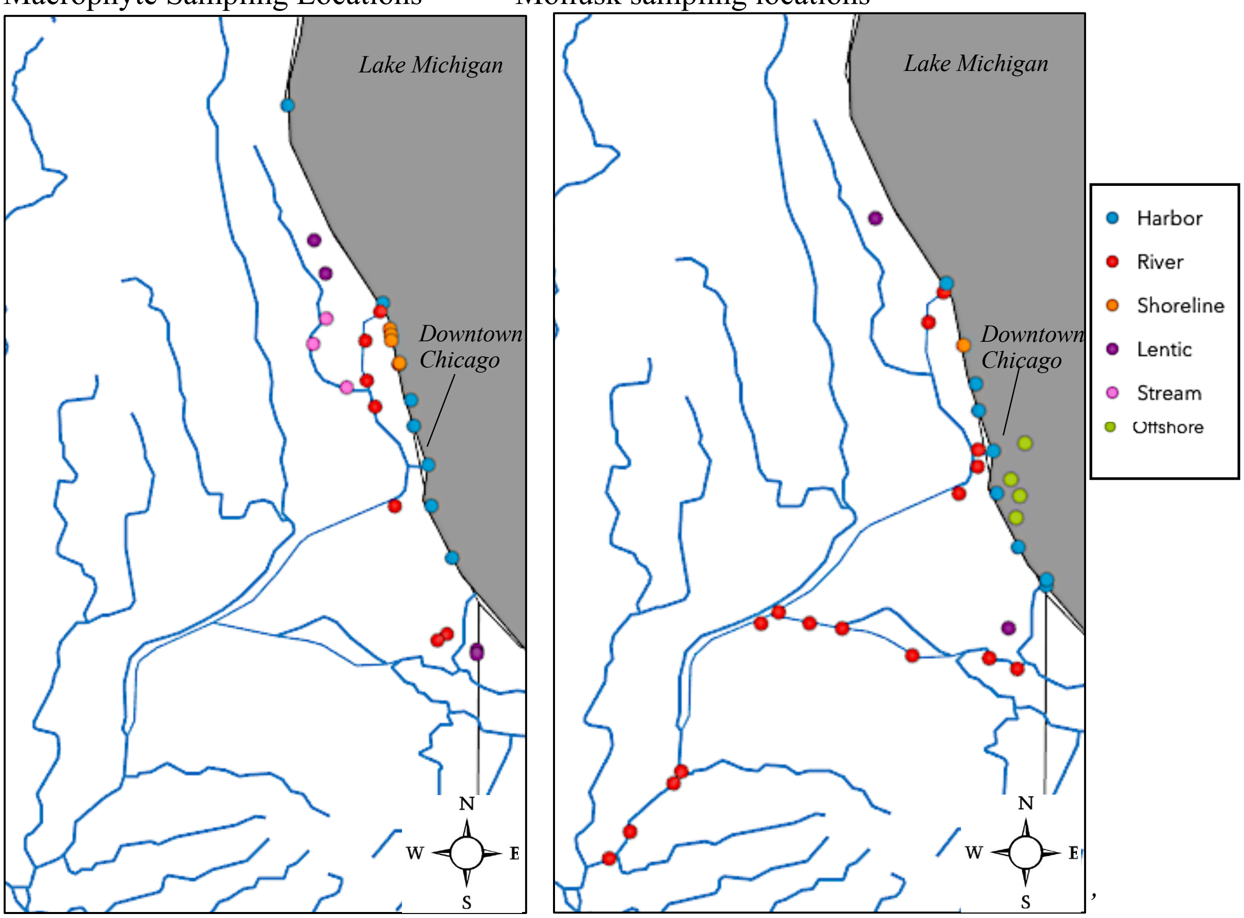


Figure 14. Sampling locations for macrophytes and mollusks. Colors indicate habitat types (see legend).

We sampled for macrophytes and mollusks in all of the harbors (N=8) to which we could gain access. There are a total of 13 harbors on the Illinois shoreline of Lake Michigan. Offshore sites (N=7) were selected based on the presence of hard substrate, such as rocky reefs and sunken ships. We focused on hard substrate because much of the remaining area of the lake is mobile sand, where we assumed that mollusks and macrophytes were considered unlikely to occur. This assumption was borne out by our sampling during which we regularly swam away from hard substrate and found zero mollusks or macrophytes on the sand.

River and lentic sampling sites were chosen to cover a wide geographical area and based upon access. Stream sites in the area are highly restricted because most lentic systems have either been filled in or greatly enlarged (Hill 2000). We sampled at three stream sites on the North Branch of the Chicago River, all of which are within the Cook County Forest Preserves.

Sampling Methods: Macrophytes

Sites were sampled for macrophytes via rake throws within harbor, shoreline, stream, lentic and river habitats. At each location, a double-sided flat-head rake attached to a rope was dropped to the bottom of the water body. The rake was dragged 2 meters along the bottom and then hauled to the surface (Deppe & Lathrop 1993; Madsen 1993; Pennsylvania Bureau of Clean Water 2015). Each rake throw therefore sampled an area 2-meters long and 40 cm wide. Once the rake was recovered we recorded total density of all macrophytes and the density of each species. The density of all macrophytes was determined by counting the total number of the tines on the rake that contained plants. Density at the species level was determined in the same way by counting the number of tines that contained each species found. A maximum of 32 tines (16 on each rake) was possible. Ten samples were taken at each location, roughly 15 meters apart.

At all sites we made 10 rake throws. In harbors, locations were often restricted because of access and we took samples haphazardly located across the area available. Samples were always taken by rake throws off docks. In rivers, rake throws were spaced 15 meters apart and were taken from a boat. Each throw occurred either close to one of the river banks or in the center of the channel. At each 15m interval we randomly chose one of the left or right bank, or the center of the channel. Stream habitats were narrower and water was always clear enough to see the bottom. These sites were sampled with the same rake throw methods as in rivers, but they were sampled while wading. At shoreline sites we visually examined areas for macrophytes. In lentic habitats, we established a transect following the shore of the lake that was 10 meters in width (Madsen 1993). Three locations along the transect were determined: along the shore, 5 meters from shore, and 10 meters from the shore. Samples were again taken every 15 meters, and sampling location within the transect was randomly chosen using the same methods as for rivers. A total of 24 sites were sampled during June – August 2016 (N=19) and June-August 2017 (N=14), across five habitat types. Nine sites were sampled during both years.

Sampling Methods: Mollusks

Harbors, lentic systems, and rivers were sampled for mollusks with Hester-Dendy samplers (HDs) during the summers of 2015 and 2017. HDs are effective for sampling epifaunal mollusks, but not burrowing species such as the Asian clam. During 2015 (N= 24 sites) and 2017 (N= 11 sites), HDs with attached weights (either tiles or bricks) were placed during June and July and remained in the water for at least six weeks (Hester & Dendy 1962). The HDs consisted of eight plates, each 7.6 cm X 7.6 cm, with varying spacing between each to allow for different sized organisms to colonize. Tiles were 15 cm X 15 cm, and bricks were standard house bricks. Two tiles or one brick were tied to the end of a rope and served as an anchor and to provide a substrate for larger organisms to colonize. Above the anchor we tied three HDs, spaced equally from the anchor to 1m below the water surface. In 2017, a tile was tied in the middle of the line to allow more surface for larger organisms to colonize. The rope was tied off to any available structure above the water surface.

Additionally, in 2015, PONAR (N= 15) and kick-net (N= 4) samples were taken from some sites in harbors, lentic, and rivers (see Table A4 for site locations). All of these samples were taken at sites where HDs were also set. In 2016, hard substrate was scraped using a D-net with the metal bar exposed. This included poles in harbors, and bridges in rivers and streams (N=10; see Table A4 for site locations). Each site was scraped at three locations, and each location scraped for one minute. Not all sites received this additional (i.e., PONAR, kick-net, scrape) sampling because many sites did not have appropriate substrate.

Mollusks were collected in a haphazard way in 2016 and 2017 from offshore sites (N=7). While SCUBA diving we hand collected mollusks when we saw them. This included samples taken from hard substrate such as rocks and shipwrecks. Results from sampling conducted using these alternative (i.e., everything except HDs) methods are reported but are not included in analyses unless otherwise stated.

We were unable to sample mollusks from stream sites. The highly variable flow patterns and high levels of public visitation made HDs unsuitable, due to the likelihood of them being damaged or stolen. Attempts to scrape with a D-net were not successful due to the lack of large hard surfaces.

All samples collected were immediately preserved in 75% ethanol for identification in the lab. Mollusks were sorted and identified to species level when possible, and each species was classified as native or non-native.

Results

Shoreline analysis

Harbors in the City of Chicago are located in close proximity to one another, with the distance between any harbor and the nearest other harbor ranging from 0.52 – 6.23 km (average = 2.93 km). The habitat between harbors largely consisted of vertical concrete or metal walls and sandy beaches. The two harbors sampled outside of the city of Chicago (Wilmette Harbor and the Great Lakes Naval Station) were located further from other harbors, but each was still less than 13 km from the nearest harbor. (See Methods: shoreline analysis for breakdown of the Illinois shoreline, by habitat type).

Macrophytes

Two non-native macrophyte species were found: curly-leaf pondweed (*Potamogeton crispus*) and Eurasian watermilfoil (*Myriophyllum spicatum*) (Figure 15). Both of these species were found in rivers, lentic systems, and harbors, but not elsewhere. Curly-leaf pondweed was never found in the absence of Eurasian watermilfoil, and was found most often in lentic systems (Table 3). Eurasian Watermilfoil was found in the highest densities in rivers (Table 3). The stream sites sampled were all located within the Forest Preserves. These sites showed no macrophyte invasion, however, they showed low rates of native organisms as well. Only two species of macrophyte were found in stream habitat, and these were both native species and at a single site (Harms Woods). These occurrences were single strands of macrophytes, covering very few tines on the rake. No macrophytes were found at the other two stream sites.

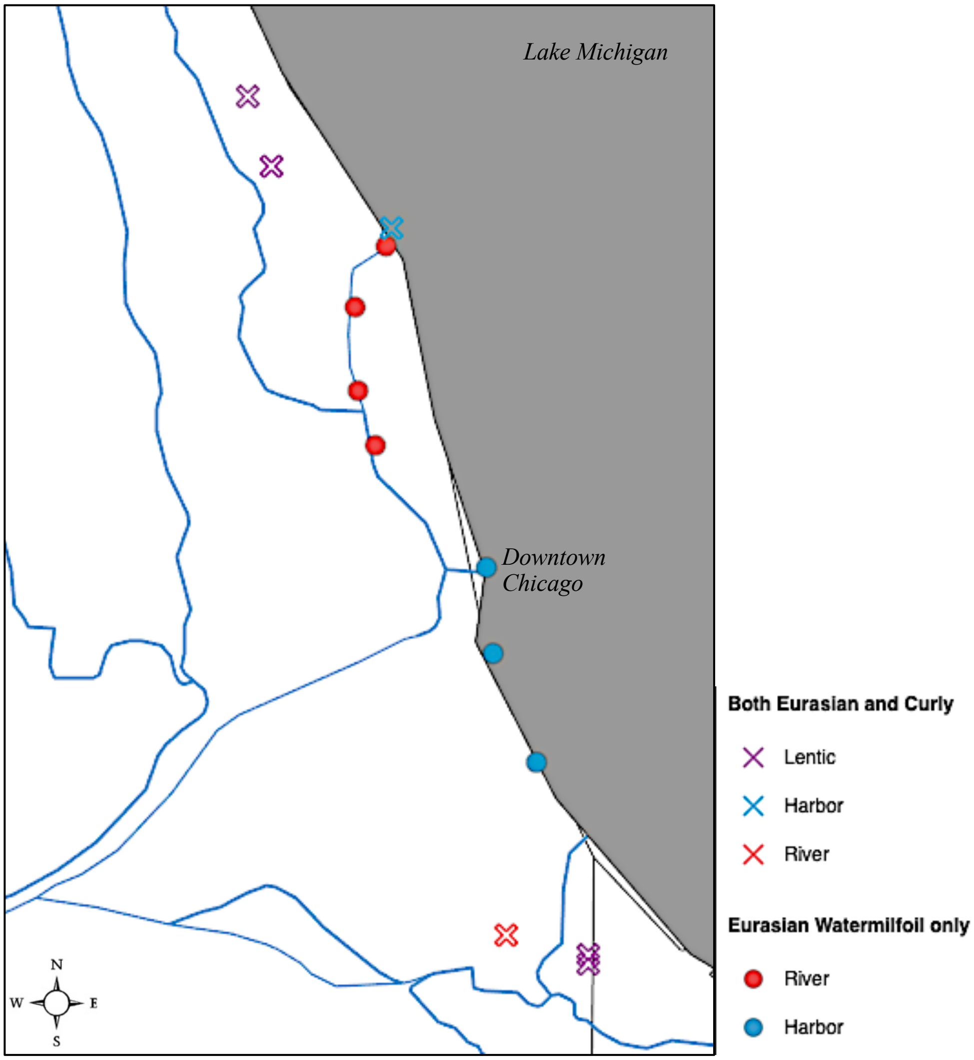


Figure 15: Location of non-native macrophyte species found during sampling in the Chicago region. Eurasian watermilfoil (*Myrophyllum spicatum*) was widespread was often found without curly-leaf pondweed (*Potamogeton crispus*). All sites with curly-leaf pondweed also had Eurasian watermilfoil.

Table 3: Species richness of macrophyte communities at each site and presence of invasive species Eurasian watermilfoil (*Myriophyllum spicatum*) and curly leaf pondweed (*Potamogeton crispus*). Number of tines covered by each species. Total number of tines for all sites was 320, except the Botanic Gardens, which the total number was 128.
Note that no macrophytes were found at offshore sites. These sites are omitted for simplicity.

		Total # Species	**Eurasian Watermilfoil**	**Curly Leaf Pondweed**
Shoreline	**Church St Pier**	0		
	Dempster St Rocks	0		
	Kedzie Rd Rocks	0		
Stream	**Labagh Woods**	0		
	Miami Woods	0		
	Harms Woods	2		
Harbor	**Montrose Harbor**	2		
	Diversey Harbor	3		
	Jackson Park Harbor	3	X (3)	
	Navy Base	3		
	31st St Harbor	4	X (8)	
	DuSable Harbor	4	X (6)	
	Wilmette Harbor	8	X (157)	X (42)
River	**Bubbly Creek**	0		
	Calumet River 1	1		
	North Shore Channel 2	2	X (92)	
	North Branch 1	3	X (31)	
	North Branch 2	4	X (7)	
	North Shore Channel 1	4	X (23)	
	Calumet River 2	6	X (2)	X (7)
Lentic	**Skokie Lagoons**	4	X (26)	X (110)
	Botanic Gardens	7	X (38)	X (8)
	Wolf Lake 1	9	X (59)	X (2)
	Wolf Lake 2	11	X (73)	X (89)

The most common native species found across rivers, lentic systems, and harbors were Elodea (*Elodea canadensis),* coontail (*Ceratophyllum demersum*)*,* and small leaf pondweed (*Potamogeton pusillus*). No invasive macrophytes were found at in the streams, and no there was an absence of all macrophytes in shoreline sites. Although we were unable to sample these habitats with the rake for practical and safety (unsafe to sample near vertical walls and rocks), we were able to visually assess these habitats and confirm that there were no macrophytes. No macrophytes were observed during our offshore SCUBA diving.

We used PERMANOVA and Non-metric Multi-Dimensional Scaling (NMDS) to examine whether there were differences in macrophyte species communities among habitat categories. We found significant differences between river, lentic systems, and harbor sites (PERMANOVA, p = 0.0337, average stress = 0.129). NMDS confirmed this and can be used to visualize the species composition data (Figure 16). Ellipses represent the standard deviation for each habitat type. Lentic and river sites were significantly different, while harbors were not significantly different to either of these.

Total macrophyte density was significantly different between lentic sites, rivers, and harbors (ANOVA, p = <0.001). Lentic sites had significantly greater density than both harbor (Tukeys HSD, p = 0.002) and river sites (Tukeys HSD, p = <0.001) (Figure 17). Harbor sites also had significantly higher density than river sites (Tukeys HSD, p = 0.04). Density of invasive species varied among sites (Table 3).

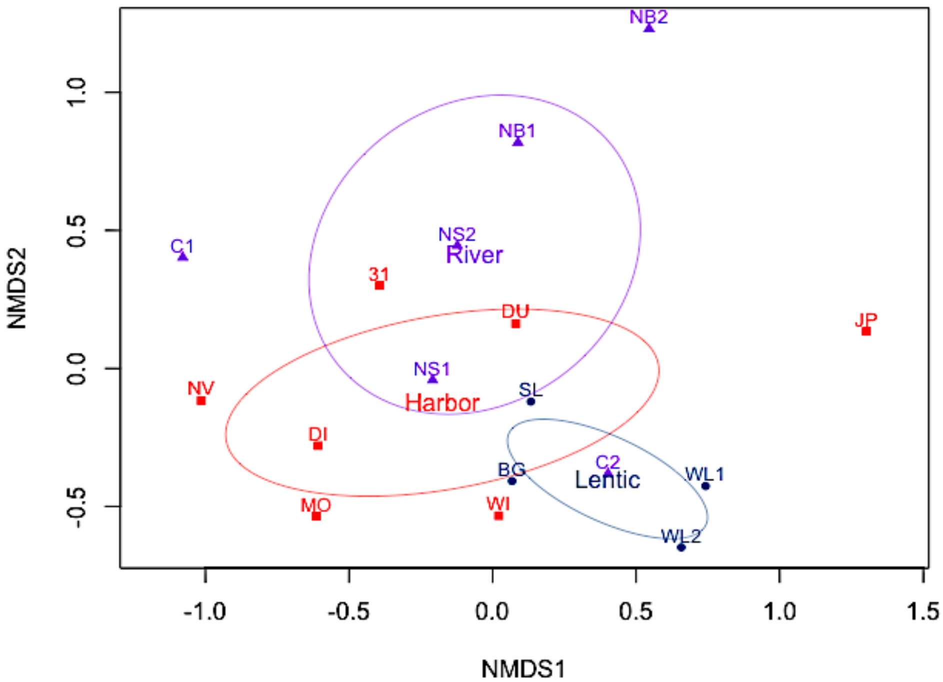


Figure 16. Two-dimensional, non-metric multidimensional scaling (NMDS) plot showing ordination of macrophyte species composition by habitat type. Average stress: 0.129. See Table A3 in appendix for site abbreviations and coordinates.

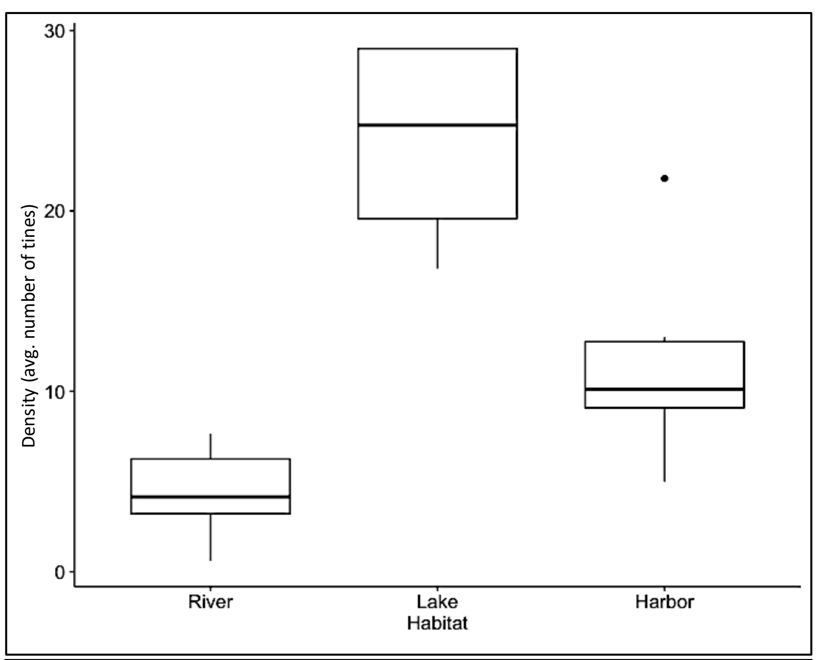


Figure 17. Total macrophyte density by habitat type. Calculated using the number of tines covered during sampling at each site.

Mollusks

Thirty-six mollusk species were identified, as well as organisms within genus *Physella* which could only be identified to this taxonomic level. Five non-native species were found during our sampling from 2015-2017: zebra mussels (*Dreissena polymorpha*), quagga mussels (*Dreissena bugensis*), a single Asian clam (*Corbicula fluminea*), faucet snails (*Bithynia tentaculata*), and a single Chinese mystery snail (*Cipangopaludina chinensis*) (Figure 18; Table 4). Both zebra and quagga mussels were commonly found throughout the region (Table 4; Figure 18a), with quagga mussels being more common. Zebra mussels were never found in the absence of quagga mussels. Faucet snails (*Bithynia tentaculata*) were found at eight river sites (Figure 18b). The Asian clam was caught while kick net sampling at site Calumet River 1 (Figure 18b). The Chinese mystery snail was a single juvenile found on a HD at site Dresden Pool 2 (Figure 18b). HDs resulted in catching four of the non-native species found: zebra mussels, quagga mussels, faucet snail, and Chinese mystery snail. Three non-native species were found in PONAR samples: zebra and quagga mussels, and faucet snails. Scraping using a D-net resulted in capturing zebra and quagga mussels. In the offshore areas of Lake Michigan, sampled via hand collecting during SCUBA, quagga mussels were the only non-native species found (Table 5). We witnessed *Dreissenid* mussels at all sites sampled via SCUBA, however, samples of these mussels were not always taken.

Table 4. Richness of mollusk community at each site and presence of invasive species zebra mussel (*Dreissena polymorpha*),), quagga mussel (*Dreissena rostriformis bugensis*), faucet snail (*Bithynia tentaculata*), and Chinese mystery snail (*Cipangopaludina chinensis*) sampled via Hester-Dendy. Note that mollusks were sampled via hand collection at offshore sites. Offshore sites are omitted for simplicity. Asian clam (*Corbicula fluminea*) was found via kick-net, this collection is omitted for simplicity

		Total # of Species	Zebra Mussel	Quagga Mussel	Faucet Snail	Chinese Mystery Snail
Shoreline	**Loyola Pier**	1		X		
Harbor	**Calumet Harbor 1**	1		X		
	Jackson Park Harbor	3		X		
	31st Street Harbor	4				
	Calumet Harbor 2	4	X	X		
	DuSable Harbor	4		X		
	Wilmett Harbor	4		X		
	Diversey Harbor	5		X		
	Montrose Harbor	10	X	X		
Lentic	**Botanic Garden**	4		X		
	Lake Calumet	5	X	X		
River	**North Shore Channel 2**	0				
	Bubbly Creek	1				
	Dresden Pool 1	2				
	Brandon Pool 1	3				
	North Branch 1	3				
	Roosevelt Bridge	3				
	Cal Sag 3	4	X	X	X	
	Dresden Pool 2	4		X		X
	Brandon Pool 2	5		X		
	North Shore Channel 1	5				
	Cal Sag 2	6	X	X	X	
	Chicago Sanitary Ship Canal 2	6	X	X	X	
	Cal Sag 1	7	X	X	X	
	Chicago Sanitary Ship Canal 1	7		X	X	
	Calumet River 1	8	X	X	X	
	Calumet River 2	12	X	X	X	

Table 5: Mollusk species found in offshore Lake Michigan sites sampled via SCUBA during 2017. Refer to Table A3 for site location coordinates.

Site	Species found
Chicago Breakwall	*Elimia livescens*
31st St. Beach	*Elimia livescens; Elimia Semicarinata*
Clemson Shoal	*Dreissena bugensis; Pleurocera canaliculata*
Monroe Break Wall	*Dreissena bugensis*
Silver Spray, Morgan Shoal	*Dreissena bugensis*
Wreck of Illinois	*Dreissena bugensis*
Wreck of Iowa	*Dreissena bugensis*

Zebra and Quagga Mussel distribution *Snails and Asian clam distribution*

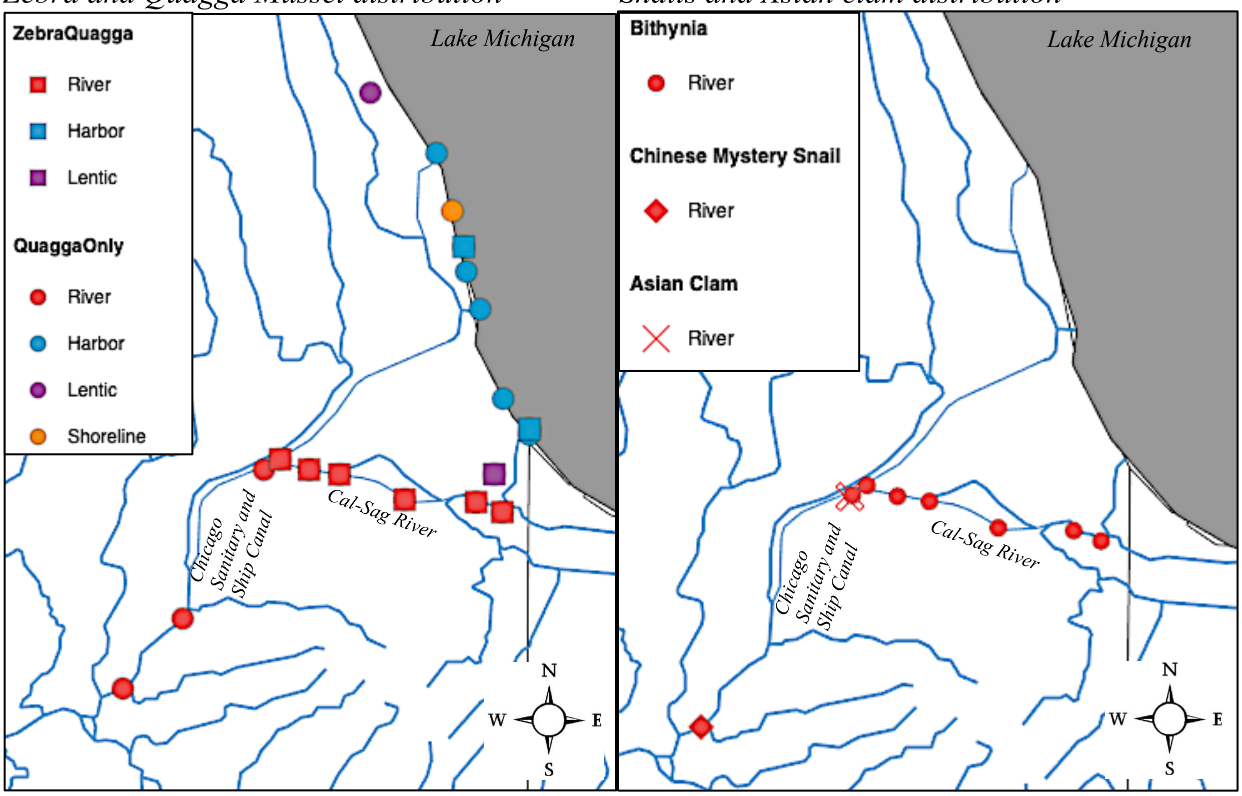


Figure 18: a) Non-native Zebra and Quagga Mussel distribution from 2015-2017. Zebra and quagga together (square), quagga only (circle). b) Non-native snail and Asian clam Hester-Dendy occurrences from 2015-2017. Faucet snail (circle), Chinese snail (diamond), Asian clam (X)

Mollusk species compositions among river, lentic, and harbor sites were not significantly different (PERMANOVA, p = 0.250, average stress = 0.133). This was confirmed by NMDS (Figure 19) where ellipses represent the standard deviation for each habitat type.

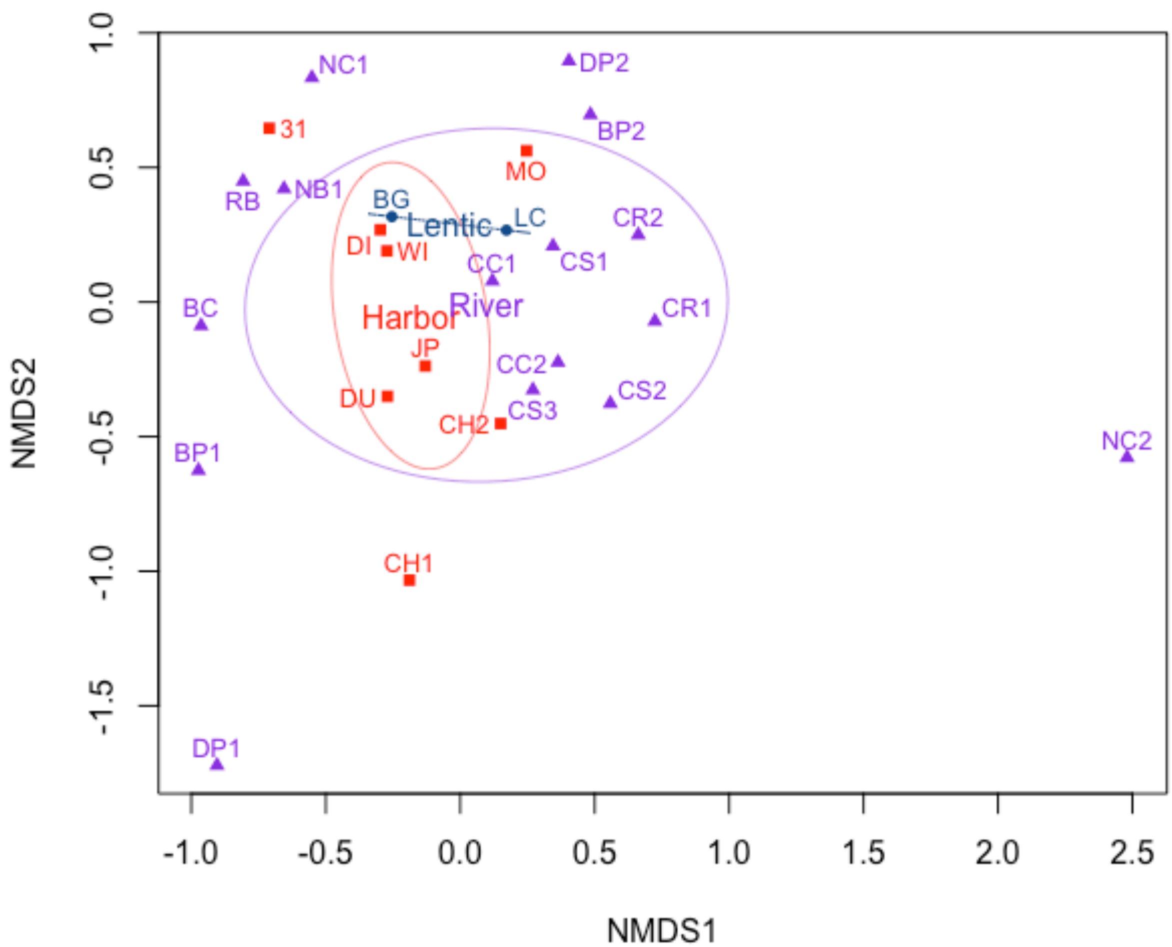


Figure 19. Two-dimensional, non-metric multidimensional scaling (NMDS) plot showing ordination of mollusk species composition by habitat type. All sites were sampled using only Hester-Dendy samplers. Average stress: 0.133. See Table A3 for site abbreviations and coordinates.

Discussion

Our analysis found that a large amount of the Illinois portion of Lake Michigan consists of altered habitat. Roughly one third (34.6%) of the shoreline is composed of artificial walls made of concrete, metal, or (very occasionally) wood. Historically, the shoreline consisted of vegetation to the water's edge and small sand beaches (Chrzastowski 1999). Beaches are now prevalent in the region and account for 27.3% of the Illinois shoreline. These have largely been formed by construction of walls perpendicular to the shoreline that trap sand. Over 13 square kilometers of new land has been formed from lakefill (Chrzastowski 1999).

Non-native macrophytes and mollusks are widespread throughout the region. Two invasive macrophyte species were found, Eurasian watermilfoil and curly-leaf pondweed (Table 3). These species were first reported in Illinois in 1916 and 1911, respectively (Jacobs & Keller 2017). Eurasian watermilfoil and curly-leaf pondweed were found in harbors, rivers, and lentic systems (Table 3). Five non-native mollusk species were found. The most widespread was the quagga mussel, which was found at 73% of the sites that we sampled by Hester-Dendy (Table 4). Quagga mussels were first reported in Illinois in 2002 (USGS NAS). Zebra mussel (first reported in Illinois in 1989) and faucet snail were also found throughout the region. One Asian clam and one Chinese mystery snail were found. Asian clams were first reported in Illinois in 1994 and Chinese Mystery Snail was first reported in 1938 (Jacobs and Keller 2017).

In marine and terrestrial systems, anthropogenically altered ecosystems have higher occurrences of non-native species compared to less altered systems (Bulleri & Airoldi 2005; Christen & Matlack 2008; Glasby et al. 2007; Gittenberger & C. van der Stelt 2011; Airoldi et al. 2015; Gaetner et al. 2016; Bieler et al. 2017). We found that this trend of increased non-native species in altered habitats is also seen in freshwater systems in the Chicago region. In our study, the forest preserves of Cook county and offshore, sandy sites served as less altered sites. The

Chicago region and the Illinois portion of Lake Michigan contain a high percentage of habitat that has been anthropogenically altered. Lotic (rivers and streams) systems in the area have been widened and hardened and connected to new waterbodies. Lentic systems are degraded from pollution, industrialization, and dredging (Hill 2000; Wilson and Weng 2010). Additionally, the creation of harbors along the Lake Michigan shoreline has increased hard substrate and created conditions that are essentially lentic in areas that previously had no such habitat. Importantly, movement of boats means that these harbors are connected by vectors which are known to transport macrophytes and mollusks (Schneider et al. 2008; Bruckerhoff et al. 2014).

The Chicago Area Waterway System (CAWS) includes the only permanent aquatic connection between the Mississippi River and Great Lakes Basins. These were not connected prior to the construction of the CSSC. All of the waterways of the CAWS have been extensively altered or are entirely artificial. During the late 1800's, canals in Chicago were built to reverse the flow of the Chicago River, causing it to flow away from Lake Michigan to improve river water quality (Hill 2000). The Chicago River, as well as other waterways in the Chicago region, have been widened, deepened, and hardened to allow for increased water flow and navigation of larger boats (Hill 2000). The area remains an active shipping hub, and cargo shipped to and through Chicago comes from the United States and locations as far as Asia and Africa (Keller et a. 2011; Chicago Maritime Museum 2019). The altered, hardened surfaces allow for the potential colonization of epifaunal mollusk species.

The City of Chicago operates 10 harbors, allowing space for 6,000 boats at one time (Chicago Harbors 2019). There are additional harbors north of the City that accommodate hundreds of additional boats. Commercial and recreational boats have the potential to travel to other harbors in the region, and travel through the Mississippi River Basin and Great Lakes Basin using the altered canals and rivers throughout the Chicago region. The developed infrastructure within harbors and along the shoreline of Lake Michigan usually consists of concrete, limestone rocks, wood, or metal replacing natural vegetation or sand (Chrzastowski 1999). Harbors alter wave exposure, possibly changing species composition (Bulleri and Chapman 2004), allowing for macrophyte species to grow in areas where they were previously not able to.

We have shown that harbors, rivers, and lentic systems in this region all contain the non-native macrophyte species Eurasian watermilfoil and curly-leaf pondweed. Eurasian watermilfoil was the most widely distributed macrophyte across the region, and at only five sites with macrophytes did we not find this species (Table 3). Three of these sites were harbors, one was the only stream site with macrophytes, and one was in the Calumet River in the south of the region. Given that Eurasian watermilfoil can have large ecosystem impacts (Jacobs and Keller 2017), these sites could be priorities for prevention by preventing transportation by humans. In particular, the harbor sites are likely able to support this species if it arrives.

Rivers and lentic systems contained different macrophyte species communities. This is not surprising due to the different characteristics of these systems such as turbidity, water flow and substrate type. We found that species composition of harbors was not significantly different from rivers or lentic systems. This indicates that species able to live in rivers but not lentic systems, and vice versa, may be able to live in harbors. The high boat traffic in harbors could result in the transportation of non-native macrophytes among harbors, as well as long distance transport to other areas of the Great Lakes Region. The shoreline areas that most resemble natural habitat (i.e. beaches, rocky areas) had no macrophytes. These habitats did contain cladophora (*Ulvophyceae sp.*), however this species is classified as an alga. We take this as an indication that historically the shoreline contained few or zero macrophytes. The high density of native and non-native macrophytes in harbors is evidence that artificial habitat allows for species to live in areas where they historically could not.

Mollusk species were rarely found in habitats that resemble the original shoreline. Attempts to scrape hard substrate (rocks) along the shoreline resulted in no mollusks being collected. Shoreline sites that did contain mollusks were highly altered to include hard substrate and consisted of piers and harbors. Non-native mollusks were common in harbors with only one harbor found to not contain invasive mollusks (Table 4). The most common species found, native or non-native, was the quagga mussel. Although not often found with Hester-Dendy samplers, our scraping of harbors collected zebra mussels in five harbors, including the single harbor were no invasive species were found using Hester-Dendy samplers. Zebra mussels were the second most common mussel species found. Many snails in the genus *Physella*, none of which are considered non-native to the Chicago region, could only be identified to genus level. The most common snail species that was classified to species was the invasive faucet snail, which was only found in river sites.

There was no difference in mollusk species composition between harbor, river, and lentic sites (Figure 19). This was possibly driven by the prevailed of quagga and zebra mussels in all habitat types. Invaders were common in sections of the CAWS that see a lot of commercial shipping, such as the Cal-Sag Channel and the Chicago Sanitary and Ship Canal (Figure 18). These artificial canals are regularly dredged (US Army Corps of Engineers 2019). Calumet Harbor and the Cal-Sag Channel carry over 14 million tons of commodities every year, 6.5 million tons also travel through the Chicago Sanitary and Ship Canal (US Army Corps of Engineers 2019). Shipping activity connects Chicago’s waterways to waterbodies throughout the United States, creating risks for the arrival and spread of many more mollusk species.

Although the habitats located between harbors do not allow for the colonization of many species, harbors are connected through boat traffic. Lake Michigan is rimmed with harbors. Both near Chicago, in Illinois, and harbors that require long-distance travel, in Indiana, Wisconsin, and Michigan. The United States portion of the Great Lakes contains over 100 harbors (US Army Corps of Engineers 2019). This indicates a substantial amount of anthropogenically altered habitat that may allow for increased establishment and rapid spread of non-native mollusk and macrophyte species. Recreational boating has aided in the spread of these species between lakes and rivers (Rothlisberger et al., 2011; Campbell et al., 2016). Boaters surveyed in Illinois were found to have visited at least two waterbodies on average within one year (Cole et al. 2019). Although most of these boaters indicated that they always clean their boats when they remove them from the water, there is still a high risk that species will be spread via recreational boating (Cole et al. 2019).

Harbors in marine systems are often hot spots for non-native species (Bulleri & Airoldi 2005; Glasby et al. 2007; Airoldi et al. 2015; Bieler et al. 2017). Hard infrastructure and artificial structures in marine systems have higher rates of non-native invertebrate species compared to natural habitat (Bulleri & Airoldi 2005; Glasby et al. 2007; Airoldi et al. 2015; Bieler et al. 2017). We found a similar pattern across our freshwater study area. Although we were unable to measure density of mollusk species, we found that non-native species are widespread in harbors in the Chicago region. These harbors are playing a similar role as rivers and inland lentic systems compared to non-anthropogenically altered shoreline habitats for non-native mollusk species. They are also aiding in the increase of macrophytes established along the coast of our study area. It is clear that anthropogenic alteration of habitat is aiding in the spread and establishment of non-native freshwater mollusks and macrophytes through the Chicago region and Lake Michigan.

CHAPTER V

CONCLUSION

The introduction and establishment of non-native species is a worldwide economic and ecological concern. In Illinois freshwaters, macrophytes, mollusks, and crayfishes are of particular concern. The Chicago region is a potential hot spot for invaders due to its large human population, role as a transportation hub, its connections to the Mississippi River Basin, and its position in the southern basin of the Laurentian Great Lakes. In this project, the role that species interactions and habitat types play in the spread of non-native species were studied. In Chapter II, the interaction between two invasive crayfish in the Chicago region was experimentally analyzed to determine which species was more successful when in competition for food and shelter. The relationship between fish and each of these species was studied to examine the role that predation may play in regulating the populations of these invasive crayfish species. *Procambarus clarkii* (red swamp crayfish), the larger of the two invasive crayfish studied (Figure 5), was not found to be more successful when directly competing with *Faxonius rusticus* (rusty crayfish) for food. However, this is likely due to the size matching and in lab experimental conditions. We believe that additional experiments without size matched crayfish would give different results. When competing for shelter, *F. rusticus* were significantly more likely to enter shelter while *P. clarkii* were significantly more likely to display aggressive behavior. In a river with a high quantity of macrophytes, *P. clarkii* were significantly more likely to be predated on, compared to *F. rusticus* (Figure 6). Chapter III addressed the lack of knowledge about the distribution of crayfishes in the Chicago region. During extensive sampling in the area we found six species of crayfish, two of which are invasive. The most common species found in the region is *F. rusticus* (Table 2; Figure 11). Another invader, *P. clarkii*, was found in high densities in the North Branch of the Chicago River and the North Shore Channel. This species was found in lower densities in harbor and lentic sites (Table 2; Figure 11). The most commonly found native species was *Faxonius virilis* which was found in multiple habitat types (Table 2; Figure 11). *Faxonius immunius* was only found in stream sites which contained relatively unaltered habitat (Table 2; Figure 11). In an offshore site within Lake Michigan, *Faxonius propinquus* were found, with no other species in the vicinity (Table 2; Figure 11). *Procambarus acutus,* a native species, was rare but found in river and stream sites. Our data was compared to data from Peters et al. (2014) to examine the change in species distributions since 1906 when the first records of crayfish in the area were made (Figure 8; Figure 11).

Beyond zebra mussels, the distribution of epifaunal mollusk and macrophyte species in the Chicago region is also poorly studied, as is the role that anthropogenic habitats play in their spread. In marine systems, artificial habitats are commonly found to contain high numbers of invasive species (Airoldi et al. 2015; Bieler et al. 2017). Chapter IV examined the distribution of habitats along the Illinois portion of the Lake Michigan shoreline, and studied the species composition of mollusks and macrophytes in each habitat type. Non-native mollusk and macrophyte species were found to be widespread throughout the region, and the most common species in each taxonomic group were invasive. Lentic and river sites were found to have different macrophyte species composition; however, harbors had a similar composition to both habitat types (Figure 16). There was no difference in epifaunal mollusk species composition between lentic, harbor, and river sites (Figure 19). Invasive mollusks were commonly found in harbors and in highly used shipping waterways. These results indicate Chicago has multiple invasive mollusk and macrophytes, and that they are most common in anthropogenically altered habitats.

This work gives important information about the current distribution of non-native and native crayfishes, mollusks, and macrophytes. Human altered freshwater habitats, such as harbors and rivers, that contain hard materials are common locations for non-native mollusk species. Harbors allow for species found in both lentic and lotic sites to colonize, increasing the likelihood that introduced species will spread throughout the Great Lakes Basin.

APPENDIX A

SUPPLEMENTARY FIGURES AND TABLES

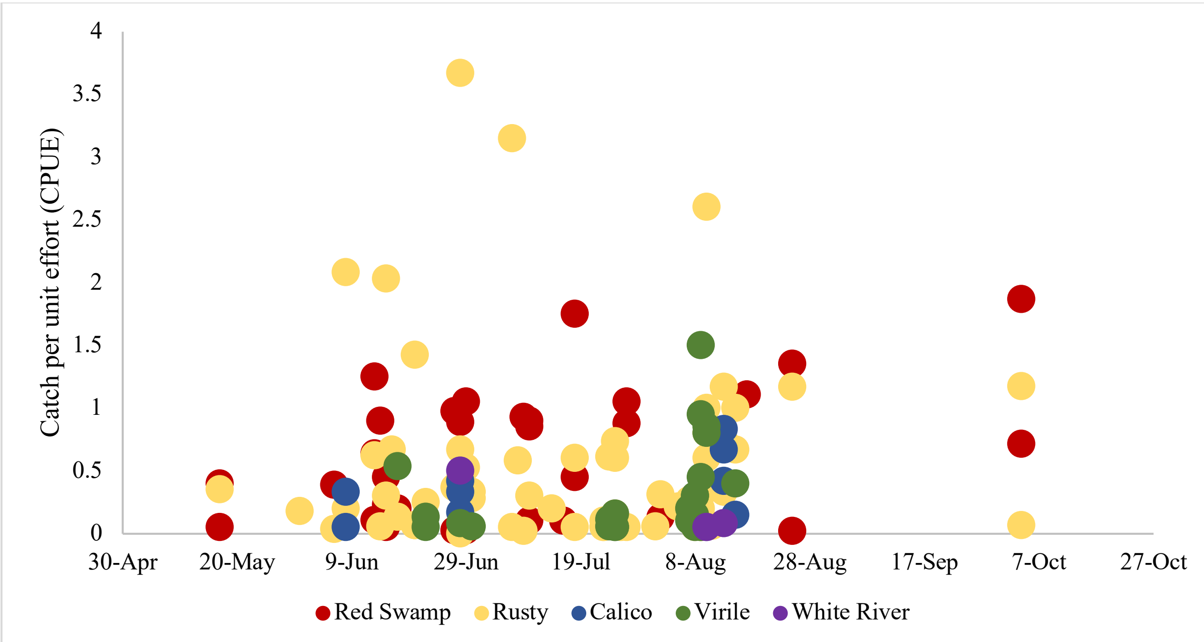


Figure A1: CPUE based on time of year sampled, showing that time of year does not have an effect on CPUE.

	Faxonius rusticus	*Procambarus clarkii*	*Faxonius virile*	*Faxonius immunius*	*Procambarus acutus*	*Faxonius propinquus*
Faxonius rusticus	River - 6 Shore - 4 Harbor - 4 Dive - 6	River - 5 Lake - 2 Harbor - 2	River - 7 Harbor - 4 Lake - 1 Stream -1 Dive - 1	River - 1 Stream - 3	River - 2 Stream - 2	0
Procambarus clarkii		Alone - 0	Lake - 1 Harbor - 1	River - 1	0	0
Faxonius virile			Lake - 7 River - 1	Stream - 1	River - 1 Stream - 1	0
Faxonius immunius				Alone - 0	0	0
Procambarus acutus					Alone - 0	0
Faxonius propinquus						Dive - 1

Figure A2: Overlap between crayfish species in the Chicago region, by habitat type.

Table A1: Table of Peters et al. (2014) Southern Lake Michigan data

Latitude	Longitude	Species	Year Found	Source
41.96434	-87.63154	*Faxonius propinquus*	1979	Quinn and Janssen 1989
42.35556	-87.82542	*Faxonius propinquus*	1980	Quinn and Janssen 1989
41.96434	-87.63154	*Faxonius virilis*	1979	Quinn and Janssen 1989
42.35556	-87.82542	*Faxonius virilis*	1980	Quinn and Janssen 1989
42.35556	-87.82542	*Faxonius propinquus*	1982	Quinn and Janssen 1989
41.96434	-87.63154	*Faxonius virilis*	1982	Quinn and Janssen 1989
42.35556	-87.82542	*Faxonius propinquus*	1983	Quinn and Janssen 1989
41.96434	-87.63154	*Faxonius virilis*	1983	Quinn and Janssen 1989
41.72694	-87.53000	*Faxonius rusticus*	1995	Kuhns and Berg 1999
41.86958	-87.61283	*Cambarus diogenes*	1906	Ortmann 1906
42.04250	-87.66989	*Cambarus diogenes*	1906	Ortmann 1906
41.86358	-87.60739	*Faxonius rusticus*	2008	University of Notre Dame
42.41071	-87.80986	*Procambarus clarkii*	2001	T Simon
42.01281	-87.66089	*Faxonius propinquus*	1979	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1979	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1980	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1980	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1982	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1982	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1983	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1983	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1990	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1990	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1992	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1992	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1994	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1994	Loyola University

Table A1: *Continued*

Latitude	**Longitude**	**Species**	**Year Found**	**Source**
42.01281	-87.66089	*Faxonius propinquus*	1995	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1995	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1996	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1996	Loyola University
42.01281	-87.66089	*Faxonius propinquus*	1998	Loyola University
42.01281	-87.66089	*Faxonius virilis*	1998	Loyola University
42.33710	-87.81910	*Unknown*	2003	Illinois Natural History Survey Lake Michigan Biological Station
42.38070	-87.80220	*Unknown*	2004	Illinois Natural History Survey Lake Michigan Biological Station
42.33710	-87.81910	*Unknown*	2005	Illinois Natural History Survey Lake Michigan Biological Station
42.21350	-87.79653	*Unknown*	2005	Illinois Natural History Survey Lake Michigan Biological Station
42.33710	-87.81910	*Faxonius virilis*	2006	Illinois Natural History Survey Lake Michigan Biological Station
42.41127	-87.78970	*Faxonius virilis*	2006	Illinois Natural History Survey Lake Michigan Biological Station
42.33710	-87.81910	*Faxonius spp.*	2007	Illinois Natural History Survey Lake Michigan Biological Station
42.48472	-87.79819	*Faxonius rusticus*	2007	Illinois Natural History Survey Lake Michigan Biological Station
42.33660	-87.80860	*Faxonius spp.*	2007	Illinois Natural History Survey Lake Michigan Biological Station
41.79220	-87.55260	*Faxonius propinquus*	2007	Illinois Natural History Survey Lake Michigan Biological Station
42.33640	-87.80411	*Faxonius rusticus*	2007	Illinois Natural History Survey Lake Michigan Biological Station
42.33710	-87.81910	*Faxonius rusticus*	2008	Illinois Natural History Survey Lake Michigan Biological Station
42.25467	-87.81692	*Faxonius spp.*	2008	Illinois Natural History Survey Lake Michigan Biological Station
42.33710	-87.81910	*Faxonius rusticus*	2008	Illinois Natural History Survey Lake Michigan Biological Station
42.33710	-87.81910	*Faxonius virilis*	2008	Illinois Natural History Survey Lake Michigan Biological Station
42.33660	-87.80860	*Faxonius propinquus*	2008	Illinois Natural History Survey Lake Michigan Biological Station
42.36156	-87.81622	*Faxonius rusticus*	2008	Illinois Natural History Survey Lake Michigan Biological Station
42.16000	-87.74670	*Faxonius propinquus*	2008	Illinois Natural History Survey Lake Michigan Biological Station
42.33640	-87.80411	*Faxonius propinquus*	2008	Illinois Natural History Survey Lake Michigan Biological Station
41.85308	-87.61003	*Faxonius rusticus*	1991	University of Vermont - E Marsden
42.36750	-87.81306	*Unknown*	2008	University of Notre Dame
42.49786	-87.80450	*Procambarus acutus*	1973	Illinois Natural History Survey

Table A1: *Continued*

Latitude	Longitude	Species	Year Found	Source
41.77658	-87.57469	*Procambarus acutus*	1975	Illinois Natural History Survey
41.87607	-87.61403	*Faxonius virilis*	1973	Illinois Natural History Survey
41.83976	-87.60619	*Faxonius virilis*	1974	Illinois Natural History Survey
41.93001	-87.63352	*Faxonius virilis*	1974	Illinois Natural History Survey
41.86040	-87.59282	*Faxonius virilis*	1984	Illinois Natural History Survey
42.01626	-87.61305	*Faxonius virilis*	1995	Illinois Natural History Survey
41.73286	-87.52950	*Faxonius virilis*	1974	Illinois Natural History Survey
42.40880	-87.80335	*Faxonius virilis*	1975	Illinois Natural History Survey
42.40880	-87.80335	*Faxonius virilis*	1980	Illinois Natural History Survey
41.77658	-87.57469	*Faxonius virilis*	1975	Illinois Natural History Survey
42.49786	-87.80450	*Faxonius virilis*	1973	Illinois Natural History Survey
41.86040	-87.59282	*Faxonius rusticus*	1984	Illinois Natural History Survey
41.92972	-87.55917	*Faxonius rusticus*	1995	Illinois Natural History Survey
41.78475	-87.57583	*Faxonius rusticus*	1995	Illinois Natural History Survey
42.30844	-87.83274	*Faxonius rusticus*	1999	Illinois Natural History Survey
41.86040	-87.59282	*Faxonius propinquus*	1984	Illinois Natural History Survey
42.40880	-87.80335	*Faxonius immunis*	1980	Illinois Natural History Survey
42.46420	-87.79900	*Faxonius immunis*	1999	Illinois Natural History Survey
42.49786	-87.80450	*Faxonius immunis*	1973	Illinois Natural History Survey
42.49786	-87.80450	*Cambarus diogenes*	1973	Illinois Natural History Survey
41.71560	-87.52769	*Faxonius rusticus*	1996	Illinois Natural History Survey
41.64240	-87.10521	*Procambarus acutus*	2002	T Simon
41.64240	-87.10521	*Faxonius virilis*	2002	T Simon
41.65675	-87.05424	*Faxonius immunis*	2002	T Simon
41.65441	-87.05995	*Faxonius immunis*	2001	T Simon

Table A2: Table of data collected from 2015-2018 in the Chicago region for this study. *Sites sampled via SCUBA

Site	Habitat Type	State	Year	# Traps	Red Swamp	Rusty	Calico	Virile	White River	Propinquus
31st St Harbor	Harbor	IL	2015	16		1				
31st St Harbor	Harbor	IL	2016	15						
31st St Harbor	Harbor	IL	2017	17		3				
31st St Harbor	Harbor	IL	2018	20		1				
59th St Harbor	Harbor	IL	2018	20						
Belmont Harbor	Harbor	IL	2018	20						
Burnham Harbor (North)	Harbor	IL	2018	18		11		2		
Burnham Harbor (South)	Harbor	IL	2018	19		14				
Diversey Harbor	Harbor	IL	2016	18		5		1		
Diversey Harbor	Harbor	IL	2017	18						
Diversey Harbor	Harbor	IL	2018	19		3		3		
DuSable Harbor	Harbor	IL	2016	12		3				
DuSable Harbor	Harbor	IL	2017	12						
Jackson Park Harbor	Harbor	IL	2015	15		2		3		
Jackson Park Harbor	Harbor	IL	2016	15	3	2		8		
Jackson Park Inner	Harbor	IL	2018	40		2		7		
Jackson Park Outer	Harbor	IL	2018	39		5		6		
Monroe Harbor	Harbor	IL	2016	3		1				
Monroe Harbor	Harbor	IL	2018	20						
Montrose Harbor	Harbor	IL	2015	15		4				
Montrose Harbor	Harbor	IL	2016	12						
Montrose Harbor	Harbor	IL	2017	16						
Montrose Harbor	Harbor	IL	2018	20		12		1		
Navy Base	Harbor	IL	2016	11						
Wilmette Harbor	Harbor	IL	2015	48		24				
Wilmette Harbor	Harbor	IL	2016	16	2	5				
Wilmette Harbor	Harbor	IL	2017	18		1				

Table A2: *Continued*

Site	Habitat Type	State	Year	# Traps	Red Swamp	Rusty	Calico	Virile	White River	Propinquus
Botanic Gardens	Lentic	IL	2017	35				2		
Calumet Lentic 1	Lentic	IL	2018	20						
Calumet Lentic 2	Lentic	IL	2018	20				1		
Jackson Lagoon East	Lentic	IL	2018	20	5	3		1		
Jackson Lagoon West	Lentic	IL	2018	20	12	1				
Skokie Lagoons 1	Lentic	IL	2017	28				2		
Skokie Lagoons 2	Lentic	IL	2018	10						
Skokie Lagoons 3	Lentic	IL	2017	20				1		
Skokie Lagoons 4	Lentic	IL	2018	10				2		
Wolf Lake 1	Lentic	IL	2015	20		4				
Wolf Lake 1	Lentic	IL	2016	20						
Wolf Lake 2	Lentic	IL	2015	20						
Wolf Lake 2	Lentic	IL	2016	18				1		
Izaak Walton League Pond	Lentic	IN	2018	20						
Marquette Park Lagoon-West	Lentic	IN	2018	20						
Marquette Park Lagoon-East	Lentic	IN	2018	20						
Wolf Lake - East	Lentic	IN	2018	20				2		
Wolf Lake - West	Lentic	IN	2018	20						
Wreck of Illinois	Offshore	IL	2016	20		1				
Wreck of Iowa	Offshore	IL	2016	20						
31st St Beach Break Wall	Offshore	IL	2017	2 ppl/20 min*		14				
Chicago Break Wall	Offshore	IL	2017	3 ppl/35 min*		4				
Clemson Shoal	Offshore	IL	2017	7 ppl/50 min*		103				
Monroe Break Wall	Offshore	IL	2017	3 ppl/35 min*		12				

Table A2: *Continued*

Site	Habitat Type	State	Year	# Traps	Red Swamp	Rusty	Calico	Virile	White River	Propinquus
Rotarian	Offshore	IL	2018	2 ppl/40 min*						
Limestone	Offshore	IL	2018	2 ppl/40 min*						
Bubbly Creek	River	IL	2015	13						25
Bubbly Creek	River	IL	2016	14						
Calumet River 1	River	IL	2015	20	2					
Calumet River 1	River	IL	2016	20		63				
Calumet River 2	River	IL	2015	20						
Calumet River 2	River	IL	2016	20						
Calumet River A	River	IL	2018	20		1		3		
Calumet River B	River	IL	2018	20		1				
Calumet River C	River	IL	2018	19						
Calumet River D	River	IL	2018	20		1		1		
Calumet River E	River	IL	2018	20						
North Branch 1 (North of River Park)	River	IL	2015	20	18					
North Branch 1 (North of River Park)	River	IL	2016	19	21					
North Branch 1 (North of River Park)	River	IL	2017	16	14					
North Branch 2 (South of River Park)	River	IL	2015	20	17					
North Branch 2 (South of River Park)	River	IL	2017	20	21	1				
North Shore Channel 1	River	IL	2015	56	52	1				
North Shore Channel 1	River	IL	2016	75	113	4				
North Shore Channel 1	River	IL	2017	181	140					
North Shore Channel 1	River	IL	2018	80	31	3				

Table A2: *Continued*

Site	Habitat Type	State	Year	# Traps	Red Swamp	Rusty	Calico	Virile	White River	Propinquus
North Shore Channel 2 (Near Wilmette)	River	IL	2017	214	18	155				
North Shore Channel 2 (Near Wilmette)	River	IL	2018	60		109				
Roosevelt Bridge	River	IL	2015	10						
Grand Calumet River-Bridge St	River	IN	2018	20		4				
Grand Calumet River- US Steel	River	IN	2018	20						
Grand Calumet River-Buchanan St	River	IN	2018	20						
Grand Calumet River-2nd Ave	River	IN	2018	20						
Grand Calumet River-Columbia Park	River	IN	2018	20		6		9		
Grand Calumet River-Peoples Park	River	IN	2018	20				19		
Grand Calumet River-Roxanna Marsh	River	IN	2018	20		20		16	1	
Grand Calumet River-Roxanna Park	River	IN	2018	20		16		17	1	
Grand Calumet River-Turner Park	River	IN	2018	20		4		30		
Little Calumet River- Oxbow	River	IN	2018	20						
Portage Burns Ditch A	River	IN	2018	20		1				
Portage Burns Ditch B	River	IN	2018	20		2				
Portage Burns Ditch C	River	IN	2018	20		1				
Portage Burns Ditch E	River	IN	2018	20						
Portage Burns Ditch F	River	IN	2018	20		2		1		
Portage Burns Ditch G	River	IN	2018	19						
Portage Burns Ditch H	River	IN	2018	19						
Church St Pier	Shore	IL	2017	15		1				
Dempster St Rocks	Shore	IL	2017	15		1				

Table A2: *Continued*

Site	Habitat Type	State	Year	# Traps	Red Swamp	Rusty	Calico	Virile	White River	Propinquus
Loyola Pier	Shore	IL	2015	16						
Loyola Pier	Shore	IL	2017	17						
Loyola Rocks	Shore	IL	2016	15		1				
Harms Woods	Stream	IL	2015	12		4	8			
Harms Woods	Stream	IL	2016	12			2			
Labagh Woods	Stream	IL	2015	12		9	5			
Labagh Woods	Stream	IL	2016	12		44	4	1	6	
Labagh Woods	Stream	IL	2017	12		25	4			
Labagh Woods	Stream	IL	2018	20		20	3			
Miami Woods	Stream	IL	2015	12		14	10		1	
Miami Woods	Stream	IL	2016	12		8	5			
Miami Woods	Stream	IL	2017	20		4	1			
Miami Woods	Stream	IL	2018	18		12				

Table A3: Site abbreviations and coordinates, sorted by habitat type.

Site	Abbreviation	Latitude	Longitude	Habitat
31st St Harbor	31	-87.60577	41.83826	Harbor
59th St Harbor	59	-87.57938	41.78727	Harbor
Belmont Harbor	BE	-87.63886	41.94320	Harbor
Burnham Harbor (North)	BUN	-87.60981	41.86417	Harbor
Burnham Harbor (South)	BUS	-87.60954	41.85649	Harbor
Diversey Harbor	DI	-87.63352	41.93226	Harbor
DuSable Harbor	DU	-87.61080	41.88653	Harbor
Jackson Park Harbor	JP	-87.57307	41.77710	Harbor
Monroe Harbor	MR	-87.61649	41.88044	Harbor
Montrose Harbor	MT	-87.63783	41.96264	Harbor
Great Lakes Naval Station	NV	-87.83286	42.30786	Harbor
Wilmette Harbor	WH	-87.68239	42.07662	Harbor
Bubbly Creek	BC	-87.66341	41.83805	River
Wolf Lake - IL Site 1	WL1	-87.53383	41.66881	Lentic
Wolf Lake - IL Site 2	WL2	-87.53387	41.66376	Lentic
Chicago Botanic Gardens	BG	-87.79080	42.15020	Lentic
Calumet Lake 1	CL1	-87.58121	41.68731	Lentic
Calumet Lake 2	CL2	-87.59588	41.67996	Lentic
Jackson Lagoon East	JLE	-87.58310	41.78790	Lentic
Jackson Lagoon West	JLW	-87.58260	41.78210	Lentic
Skokie Lagoons - Site 1	SL1	-87.77501	42.11392	Lentic
Skokie Lagoons - Site 2	SL2	-87.77283	42.11113	Lentic
Skokie Lagoons - Site 3	SL3	-87.76953	42.11433	Lentic
Skokie Lagoons - Site 4	SL4	-87.77071	42.11549	Lentic
Izaak Walton League Pond	IZ	-87.16827	41.61216	Lentic
Little Calumet River - Oxbow	LC	-87.45670	41.57169	Lentic

Table A3: *Continued*

Site	Abbreviation	Latitude	Longitude	Habitat
Marquette Park Lagoon - West	MPW	-87.27220	41.61480	Lentic
Marquette Park Lagoon - East	MPE	-87.26060	41.61820	Lentic
Wolf Lake - IN W	WLE	-87.52339	41.67366	Lentic
Wolf Lake - IN E	WLW	-87.51348	41.66867	Lentic
Calumet River - Site 1	CR1	-87.61713	41.65054	River
Calumet River - Site 2	CR2	-87.57433	41.63835	River
Calumet River A	CRA	-87.56251	41.64610	River
Calumet River B	CRB	-87.58901	41.64305	River
Calumet River C	CRC	-87.60834	41.65024	River
Calumet River D	CRD	-87.62092	41.65860	River
Calumet River E	CRE	-87.63338	41.65748	River
Calumet River F	CRF	-87.65682	41.65240	River
North Branch - Site 1	NB1	-87.69450	41.95497	River
North Branch - Site 2	NB2	-87.70760	41.98559	River
North Shore Channel - Site 1	NC1	-87.70983	42.03238	River
North Shore Channel - Site 2	NC2	-87.68644	42.06650	River
Roosevelt Bridge	RB	-87.63462	41.86863	River
Grand Calumet River - Bridge	GC1	-87.37238	41.60925	River
Grand Calumet River - US Steel	GC2	-87.33435	41.60766	River
Grand Calumet River - Buchanan	GC3	-87.34759	41.60954	River
Grand Calumet River - 2nd Ave	GC4	-87.35888	41.60722	River
Grand Calumet River - Columbia	GC5	-87.49914	41.61827	River
Grand Calumet River - Peoples	GC6	-87.51354	41.62255	River
Grand Calumet River - Roxanna Marsh	GC7	-87.48799	41.61802	River
Grand Calumet River - Roxanna Park	GC8	-87.49064	41.61604	River
Grand Calumet River - Turner	GC9	-87.51289	41.62250	River
Portage-Burns Ditch A	PBA	-87.17629	41.63357	River

Table A3: *Continued*

Site	**Abbreviation**	**Latitude**	**Longitude**	**Habitat**
Portage-Burns Ditch B	PBB	-87.17697	41.63116	River
Portage-Burns Ditch C	PBC	-87.17665	41.62341	River
Portage-Burns Ditch D	PBD	-87.18301	41.61510	River
Portage-Burns Ditch E	PBE	-87.17613	41.61524	River
Portage-Burns Ditch F	PBF	-87.17398	41.61250	River
Portage-Burns Ditch G	PBG	-87.15952	41.61138	River
Portage-Burns Ditch H	PBH	-87.18021	41.60982	River
Church Street Pier	CS	-87.67144	42.04609	Shoreline
Dempster Street Rocks	DE	-87.66950	42.03918	Shoreline
Kedzie Street Rocks	KE	-87.66908	42.03257	Shoreline
Loyola Beach	LB	-87.65701	42.00462	Shoreline
Loyola Pier	LP	-87.65651	42.00632	Shoreline
Loyola Rocks	LR	-87.65582	42.00021	Shoreline
Harms Woods	HW	-87.77158	42.05838	Stream
LaBaugh Woods	LA	-87.73928	41.97762	Stream
Miami Woods	MW	-87.79222	42.02867	Stream
Chicago Breakwall	CBW	-87.59356	41.89685	Offshore
Clemson Shoal	CM	-87.54559	41.80761	Offshore
Monroe Breakwall	MBW	-87.59025	41.88281	Offshore
Silver Spray/Morgan Sholes	SS	-87.57618	41.81055	Offshore
Wreck of Illinois	IL	-87.57050	41.83549	Offshore
Wreck of Iowa	IW	-87.56251	41.89522	Offshore
31st St Beach & Breakwall	31B	-87.36216	41.50406	Offshore
Rotarian	RO	-87.43443	41.95027	Offshore
Limestone Bed	LM	-87.48066	41.95031	Offshore

Table A4: Locations of kicknet, ponar, and scraping sampling for mollusks in 2015 and 2016.

Site	Habitat	Year Sampled	Method
Calumet Harbor 1	Harbor	2015	Kicknet
Calumet Harbor 2	Harbor	2015	Kicknet
Montrose Harbor	Harbor	2015	Kicknet
Wilmette Harbor	Harbor	2015	Kicknet
31st Street Harbor	Harbor	2015	Ponar
Montrose Harbor	Harbor	2015	Ponar
Brandon Road Pool 1	River	2015	Ponar
Brandon Road Pool 2	River	2015	Ponar
Bubbly Creek	River	2015	Ponar
Cal-Sag River 2	River	2015	Ponar
Cal-Sag River 3	River	2015	Ponar
Calumet River 1	River	2015	Ponar
Calumet River 2	River	2015	Ponar
Dresden Pool 1	River	2015	Ponar
Dresden Pool 2	River	2015	Ponar
Little Calumet River	River	2015	Ponar
North Shore Channel 1	River	2015	Ponar
North Shore Channel 2	River	2015	Ponar
Roosevelt Bridge - Main Branch	River	2015	Ponar
31st Street Harbor	Harbor	2016	Scraping
DuSable Harbor	Harbor	2016	Scraping
Jackson Park Harbor	Harbor	2016	Scraping
Montrose Harbor	Harbor	2016	Scraping
Wilmette Harbor	Harbor	2016	Scraping
Bubbly Creek	River	2016	Scraping
Little Calumet River	River	2016	Scraping
Harms Woods	Stream	2016	Scraping
Labagh Woods	Stream	2016	Scraping
Miami Woods	Stream	2016	Scraping

LITERATURE CITED

Airoldi, L., Turon, X., Perkol-Finkel, S., Ruis, M. (2015) Corridors for aliens but not for natives: effects of marine urban sprawl at a regional scale. Diversity and Distributions 21: 755 768

Anastácio, P.M. and Marques, J.C. (1997) Crayfish, Procambarus clarkii, effects on initial stages of rice growth in the lower Mondego River valley (Portugal). Freshwater Crayfish 11: 608-617

Anastácio, P.M., Correia, A.M., Menino, J.P., da Silva, M. (2005) Are rice seedlings affected by changes in water quality caused by crayfish? International Journal of Limnology 41(1): 16

Albertson, L.K. and Daniels, M.D. (2016) Effects of invasive crayfish on fine sediment accumulation, gravel movement, and macroinvertebrate communities. Freshwater Science 35: 644-653

Alonso, F., Martínez, R. (2006) Shelter competition between two invasive crayfish species: a laboratory study. Bulletin François de la Peche et de la Pisciculture 380-381: 1121-1132

ArcGIS ESRI v 10.6.1 (2018)

Barbaresi, S., Tricarico, E., Gherardi, F. (2004) Factors inducing the intense burrowing activity by the red swamp crayfish, *Procambarus clarkii*, an invasive species. The Science of Nature 91(7): 342-345

Bax N., Williamson A., Aguero M., Gonzalez E., Geeves W. (2003) Marine invasive alien species: a threat to global biodiversity. Marine Policy 313-323

Behringer, D.C. and Hart, J.E. (2017) Competition with stone crabs drives juvenile spiny lobster abundance and distribution. Oecologia 184: 205-218

Behm, J. (2009) Bleach set to eradicate Germantown's invasive crayfish. Milwaukee, Wisconsin Journal Sentential

Bellard, C., Thuiller, W., Leroy, B., Genovesi, P., Bakkenes, M., Courchamp, F. (2013) Will climate change promote future invasions? Global Change Biology 19(12): 3740-3748

Bieler, R., Grandados-Cifuentes, C., Rawlings, T.A., Sierwald, P., Collins, T.M. (2017) Non native molluscan colonizers on deliberately placed shipwrecks in the Florida Keys, with description of a new species of potentially invasive worm-snail (Gastropoda: Vermetidae). PeerJ 5: e3158

Bruckerhoff. L., Havel, J., Knight, S. (2014) Survival of invasive aquatic plants after air exposure and implications for dispersal by recreational boats. Hydrobiologia 746: 113-121

Bulleri, F., and Chapman, M.G. (2004) Intertidal assemblages on artificial and natural habitats in marinas on the north-west coast of Italy. Marine Biology 145(2): 381-391

Bulleri, F. and Airoldi, L. (2005) Artificial Marine Structures Facilitate the Spread of a Non Indigenous Green Alga, *Codium fragile* ssp. *tomentosoides*, in the North Adriatic Sea. Journal of Applied Ecology 42(6): 1063-1072

Bunk, H. (2014) Containment, Control and Eradication of Ambitious Architect: *Procambarus clarkii*, The Red Swamp Crayfish

Bunn, S.E. and Arthington, A.H. (2002) Basic Principles and Ecological Consequences of Altered Flow Regimes for Aquatic Biodiversity. Environmental Management 30(4): 492-507

Butler, M. and Stein, R. (1985) An analysis of the mechanisms governing species replacement in crayfish. Oecologia 66: 168-177

Campbell, T., Verboomen, T., Montz, G., Seilheime, T. (2016) Volume and contents of residual water in recreational watercraft ballast systems. Management of Biological Invasions 7(3): 281-286

Capelli, G.M. and Magnuson, J.J. (1983) Morphoedaphic and biogeographic analysis of crayfish distribution in Northern Wisconsin. Journal of Crustacean Biology 3: 548-564

Carpenter, S.R. & Lodge, D.M. (1986) Effects of Submersed Macrophytes on Ecosystems Processes. Aquatic Botany 26: 341-370

Chicago Harbors (2019) Explore the Harbors. https://www.chicagoharbors.info/harborspage/

Chicago Maritime Museum (2019) History http://www.chicagomaritimemuseum.org/history/

Childress, M.J. and Herrnkind, W.F. (1994) The behavior of Caribbean juvenile spiny lobster in Florida Bay: seasonality, ontogeny and sociality. Bulletin of Marine Science 54: 819 827

Christen, D.C. and Matlack, G.R. (2009) The habitat and conduit functions of roads in the spread of three invasive plant species. Biological invasions 11(2): 453-465

Chucholl, C. and Wendler, F. (2016) Positive selection of beautiful invaders: long-term persistence and bio-invasion risk of freshwater crayfish in the pet trade. Biological Invasions 19(1): 197-208

Chrzastowski, M (1999) Department of Natural Resources, Illinois State Geological Survey, Geology of the Chicago Lakeshore: Shaping the Chicago Lakeshore (poster)

Clark, J.M., Kershner, M.W., Montemarano, J.J. (2013) Habitat-specific effects of particle size, current velocity, water depth, and predation risk on size-dependent crayfish distribution. Hydrobiologica 716: 103-114

Clarke Murray, C., Gartner, H., Gregr, E.J., Chan, K., Pakhomov, E., Therriault, T.W. (2014) Spatial distribution of marine invasive species: environmental, demographic and vector drivers. Diversity and Distributions 20: 824-836.

Clinton, W. J. (1999) Presidential Documents, Executive Order 13112, Invasive Species. *Federal Register,* 64, 6183-6186.

Cole, E., Keller, R.P., Garbach, K. (2019) Risk of invasive species spread by recreational boaters remains high despite widespread adoption of conservation behaviors. Journal of Environmental Management 229: 112-119

Crall A.W., Newman, G.J., Stohlgren, T.J., Jarnevich, C.S., Evangelista, P., Guenther, D. (2006) Evaluating dominance as a component of non-native species invasions. Diversity and Distributions 12: 195-204

Crandall, K.A. and De Grave, S. (2017) An updated classification of the freshwater crayfishes (Decapoda: Astacidea) of the world, with a complete species list. Journal of Crustacean Biology 37: 615-653

Cruz, M.J. and Rebelo, R. (2006) Colonization of freshwater habitats by an introduced crayfish, *Procambarus clarkii*, in Southwest Iberian Peninsula. Hydrobiologica 575: 191-201

Deppe, E.R. & Lathrop, R.C. (1993) Recent changes in the aquatic macrophyte community of Lake Mendota. Transactions 81: 41-58

DiDonato, G.T. and Lodge, D.M. (1993) Species Replacements among *Orconectes* Crayfishes in Wisconsin Lakes: The Role of Predation by Fish. Canadian Journal of Fisheries and Aquatic Science 50: 1484-1488.

DiStefano, R.J., Reitz, R.A., Imhoff, E.M. (2016) Examining One State's Regulation Development Process to Manage Alien Crayfish Introductions. Fisheries 41(12): 726-737

Donato, R., Rollandin, M., Favaro, L., Ferrarese, A., Pessani, D., Ghia, D. (2018) Habitat use and population structure of the invasive red swamp crayfish Procambarus clarkii (Girard, 1852) in a protected area in northern Italy. Knowledge & Management of Aquatic Ecosystems 419: 12

Dudgeon D., Arthington A.H., Gessner, M.O., Kawabata, Z., Knowler, D.J., Leveque, C., Naiman, R.J., Prieur-Richard, A., Soto, D., Stiassny, M.L.J, Sullivan, C.A. (2005) Biological Reviews 81:163-182

Drenner, R.W., Gallo K.L., Edwards, C.M., Rieger, K.E., Dibble, E.D. (1997) North American Journal of Fisheries Management 17(4): 1010-1013

Drake J.M. and Bossenbroek, J.M. (2004) The potential distribution of zebra mussels in the United States. BioScience 54: 931-941

Edgerton B.F., Evans, L.H., Stephans, F.J., Overstreet, R.M. (2002) Synopsis of Freshwater Crayfish Diseases and Commensal Organisms. Aquaculture 206(1-2): 57-135

Englund, C. & Krupa, J.J. (2000) Habitat use by crayfish in stream pools: influence of predators, depth and body size. Freshwater Biology 43: 75-83

Fitzsimons, J.D., Jonas, J.L., Claramunt, R.M., Williston, B., Williston, G., Marsden, J.E., Ellrott, B.J., Honeyfield, D.C. (2007) Influence of egg predation and physical disturbance on lake trout *Salvelinus namaycush* egg mortality and implications for life-history theory. Journal of Fisheries Biology 71: 1–16

Fletcher, RJ Jr (2007) Species Interactions and Population Density Mediate the Use of Social Cues for Habitat Selection. Journal of Animal Ecology 76: 598-606Gherardi, F,

Gaertner, M., Larson, B.M.H., Irlich, U.M., Holmes, P.M., Stafford, L., van Wilgen, B.W., Richardson, D.M. (2016) Managing invasive species in cities: A framework from Cape Town, South Africa. Landscape and Urban Planning 151: 1-9

Gherardi, F., Tricarico, E., Ilheu, M. (2002) Movement patterns of an invasive crayfish, *Procambarus clarkii*, in a temporary stream of southern Portugal. Ethology Ecology & Evolution (14) 183-197

Gherardi, F., Daniels, W.H. (2004) Agonism and shelter competition between invasive and indigenous crayfish species. Canadian Journal of Zoology 82: 1923-1932

Gherardi, F. (2010) Invasive Crayfish and Freshwater Fishes of the World. Revue scientifique et technique 29(2): 241-254

Gherardi, F., Aquiloni, L, Diéguez-Uribeondo, J., Tricarico, E. (2011) Managing invasive crayfish: Is there a hope? Aquatic Sciences 73(2): 185-200

Gittenberger, A. & C. van der Stelt, R. (2011) Artificial structures in harbors and their associated ascidian fauna. Aquatic Invasions 6(4): 413-420

Glasby, T.M., Connell, S.D., Holloway, M.G., Hewitt, C.L (2007) Nonindigenous biota on artificial structures: could habitat creation facilitate biological invasions? Marine Biology 151(3) 887-895

Glon, M., Larson, E.R., Reisinger, L.S., Pangle, K.L. (2017) Invasive dreissenid mussels benefit invasive crayfish but not native crayfish in the Laurentian Great Lakes. Journal of Great Lakes Research 43(2): 289-297

Google Earth v 7.3.2 (2018) Illinois portion of the Lake Michigan shoreline

Great Lakes Integrated Science Assessments (GLISA) (2014) Climate change in the Great Lakes Region.http://glisa.umich.edu/media/files/GLISA_climate_change_summary.pdf

Groves, R. (1991) A short history of biological invasion in Australia Biogeography of Mediterranean invasion, Cambridge University Press, Cambridge, UK, 59-63

Harvey, G.L., Moorhouse, T.P., Clifford, N.J., Henshaw, A.J., Johnson, M.F., Macdonald, D.W., Reid, I., Rice, S.P. (2011) Evaluating the role of invasive aquatic species as drivers of fine sediment-related river management problems: The case of the signal crayfish (*Pacifastacus leniusculus*). Progress in Physical Geography 35(4): 517–533

Havel, J.E., Kovalenko, K.E., Tomaz, S.M., Amalfitano, S., Kats, L.B. (2015) Aquatic invasive species: challenges for the future. Hydrobiologica 750:147–170

Hein, C.L., Roth, B.M, Ives, A.R., Vander Zanden, M.J. (2006) Fish predation and trapping for rusty crayfish (*Orconectes rusticus*) control: a whole-lake experiment. Canadian Journal of Fisheries and Aquatic Science 63: 383–393

Hein, C.L., Vander Zanden, M.J., Magnuson, J.J. (2007) Intensive trapping and increased fish predation cause massive population decline of an invasive crayfish. Freshwater Biology 52: 1134-1146

Hill, A.M. and Lodge, D.M.(1999) Replacement of Resident Crayfishes by an Exotic crayfish: the Roles of Competition and Predation. Ecological Applications 9(2): 678-690

Hill, L. (2000) The Chicago River: A Natural and Unnatural History. Southern Illinois University Press

Hoffman, J.C., Schlosser, J., Trebitz, A.S., Peterson, G.S., Gutsch, M., Quinlan, H., Kelly, J.R. (2015) Sampling Design for Early Detection of Aquatic Invasive Species in Great Lakes Ports. Fisheries 41(1): 26-37

Issa, F.A., Adamson, D.J., Edwards, D.H. (1999) Dominance hierarchy formation in juvenile crayfish Procambarus clarkii. Journal of Experimental Biology 202: 3497-3506

Jacobs, A.I., and Keller, R.P (2017) Straddling the divide: invasive aquatic species in Illinois and movement between the Great Lakes and Mississippi basins. Biological Invasions 19: 635-646

Janssen, J.J., Berg, M.B., Lozano, S.J., 2005. Submerged terra incognita: Lake Michigan's abundant but unknown rocky zones. In: Edsall, T., Munawar, M. (Eds.), The State of Lake Michigan: Ecology, Health and Management. Ecovision World Monograph Series.

Johnson, L.E., Ricciardi, A., Carlton, J.T. (2001) Overland dispersal of aquatic invasive species: a risk assessment of transient recreational boating. Ecol Appl 11:1789–1799

Johnson, P.T.J., Olden, J.D., Solomon, C.T., Vander Zanden, M.J. (2009). Interactions among invaders: community and ecosystem effects of multiple invasive species in an experimental aquatic system. Oecologia 159:161–170

Jonas, J.L., Claramunt, R.M., Fitzsimons, J.D., Marsden, J.E., Ellrott, B.J., 2005. Estimates of egg deposition and effects of lake trout (Salvelinus namaycush) egg predators in three regions of the Great Lakes. Canadian Journal of Fish and Aquatic Science 62, 2254–2264.

Karatayev, A.Y., Burlakova, L.E., Padilla, D.K. (2014) Zebra versus quagga mussels: a review of theirspread, population dynamics, and ecosystem impacts. Hydrobiologica 746: 97-112

Karavanich, C. and Atema, J. (1998) Individual recognition and memory in lobster dominance. AnimalBehavior 56: 1553–1560

Keller, R.P. and Lodge D.M (2007) Species invasions from commerce in live aquatic organisms: problems and possible solutions. BioScience 57: 428-436.

Keller, R.P., Drake, J.M., Drew, M.B., & Lodge, D.M. 2011. Linking environmental conditions and ship movements to estimate invasive species transport across the global shipping network. *Diversity and Distributions*, 17, 93-102.

Keller R.P., Masoodi, A., Shackleton, R.T. (2018) The impact of invasive aquatic plants on ecosystem services and human well-being in Wular Lake, India. Regional Environmental Change 18: 847-857.

Klocker, C.A. and Strayer, D.L. (2004) Interactions Among an Invasive Crayfish (*Orconectes rusticus*), a Native Crayfish (*Orconectes limosus*), and Native Bivalves (Sphaeriidae and Unionidae). Northeastern Naturalist 11: 167-178

Kolar C.S., Lodge D.M. (2001) Progress in invasion biology: predicting invaders. Trends in Ecology and Evolution 16: 199-204

Kovalenko, K.E. & Dibble, E.D (2014) Invasive macrophyte effects on littoral trophic structure and carbon sources. Hydrobiologica 721: 23-34

Kreps T.A., Larson E.R., Lodge D.M. (2016) Do invasive rusty crayfish (Orconectes rusticus) decouple littoral and pelagic energy flows in lake food webs? Freshwater Science 35: 103-113.

Lodge D.M., Kershner M.W., Aloiant J.E., Covich A.P. (1994) Effects of an omnivorous crayfish (Orconectes rusticus) on a freshwater littoral food source. Ecology 75: 1265 1281

Lodge, D.M., Deines, A., Gherardi, F., Yeo, D.C.J., Arcella, T., Baldridge, A.K., Barnes, M.A., Chadderton, W.L., Feder, J.L., Gantz, C.A., Howard, G.W., Jerde, C.L., Peters, B.W., Peters, J.A., Sargent, L.W., Turner, C.R., Wittmann, M.E., Zeng, Y. (2012) Global Introductions of Crayfishes: Evaluating the Impact of Species Invasions on Ecosystem Services. Annual Review of Ecology Evolution and Systematics 43(1):449-472

Loureiro, T., Anastácio, P.M., Bueno, S.L.S., Araujo, P.B. (2015) Distribution, introduction pathway, and invasion risk analysis of the North American crayfish Procambarus clarkii (Decapoda: Cambaridae) in southeast Brazil. Journal of Crustacean Biology 35(1):88

Macisaac, H.J. (1996) Potential abiotic and biotic impacts of zebra mussels on the inland waters of North America. American Zoologist (36) 287-299.

Madsen, J. D. (1993) Biomass techniques for monitoring and assessing control of aquatic vegetation. Lake and Reservoir Management 7: 141-154

Martel, A.L. and Maddill, J.B. (2018) Twenty-six years (1990–2015) of monitoring annual recruitment of the invasive zebra mussel (*Dreissena polymorpha*) in the Rideau River, a small river system in Eastern Ontario, Canada. Candian Journal of Zoology 96: 1071-1079

Matthews, J., Van der Velde, G., Bij de Vaatea, A., Collar, F.P.L., Koopman, K.R., & Leuven, R.S.E.W. (2014). Rapid range expansion of the invasive quagga mussel in relation to zebra mussel presence in The Netherlands and Western Europe. *Biological Invasions*, 16, 23-42.

Mazlum, Y. (2007) Effects of temperature on the survival and growth of two cambarid crayfish juveniles. Crustaceana 80:947-954

McKinney, M.L. (2006) Urbanization as a major cause of biotic homogenization. Biological Conservation 127(3): 247-260

Mills, E.L., Leach, J.H., Carlton, J.T., Secor, C.L. (2004) Exotic species and the integrity of the Great Lakes. Bioscience 44: 666-676

Myers, J.H., Simbrloff, D., Kuris, A.M., Carey, J.R. (2000) Eradication revisited: dealing with exotic species. Trends in Ecology and Evolution 15(8): 316-320

Nilsson, C., Reidy, C.A., Dynesuis, M., Revenga, C. (2005). Fragmentation and Flow Regulation of the World's Large River Systems. Science 308(5720): 405-408

Nunes, A.L., Hoffman, A.C., Zengeya, T.A., Measey, G.J., Weyl, O.L.F. (2017) Red swamp crayfish, *Procambarus clarkii*, found in South Africa 22 years after attempted eradication. Aquatic Conservation 27: 1334-1340.

Oreska, M.P.J. and Aldridge, D.C. (2011) Estimating the financial costs of freshwater invasive species in Great Britain: a standardized approach to invasive species costing. Biological Invasions 13: 305-319 104

Pavey, C. R. and Fielder, D. R. (1996). The influence of size differential on agonistic behaviour in the freshwater crayfish, *Cherax cuspidatus* (Decapoda: Parastacidae). Journal of Zoology London 238: 445-457

Pejchar, L. and Mooney, H.A. (2009) Invasive species, ecosystem services and human well 105
being. Trends in Ecology & Evolution 24(9): 497-504.

Pennsylvania Bureau of Clean Water (2015) Aquatic macrophyte coverage procedures for lake assessments.

Peters, J.A.,Cooper, M.J., Creque, S.M., Kornis, M.S., Maxted, J.T., Pretty, W.L., Schueler, F.W., Simon, T.P., Taylor, C.A., Thoma, R.F., Uzarski, D.G., Lodge, D.M. (2014) Historical changes and current status of crayfish diversity and distribution in the Laurentian Great Lakes. Journal of Great Lakes Research 40(1): 35-46

Pimentel, D., Zuniga, R., Morrison, D. (2005) Update on the environmental and economic costs associated with alien-invasive species in the United States. Ecological Economics 52(3): 273-288

Quinn, J.P., Janssen, J., (1989) Crayfish competition in southwestern Lake Michigan: a predator mediated bottleneck. Journal of Freshwater Ecology 5 (1), 75–85

R Core Development Team (2015) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria

Rabeni, C.F. (1985) Resource Partitioning by Stream-dwelling Crayfish: The Influence of Body Size. The American Midland Naturalist 113: 20-29.

Redman, R., Mackey, S., Dub, J., Czesny, S. (2017) Lake trout spawning habitat suitability at two offshore reefs in Illinois waters of Lake Michigan. Journal of Great Lakes Research 43: 335-344

Ricciardi, A. and Rasmussen, J.B. (1999) Extinction Rates of North American Freshwater Fauna. Conservation Biology 13: 1220-1222

Ricciardi, A. (2006) Patterns of invasion in the Laurentian Great Lakes in relation to changes in vector activity. Diversity Distributions (12) 425-433

Ricciardi, A., Palmer, M.E., Yan, N.D. (2011) Should Biological Invasions Be Managed as NaturalDisasters? BioScience 61: 312-317

Rodríguez, C.F., Becares, E., Fernandez-Alaez, M. (2003) Shift from clear to turbid phase in Lake Chozas (NW Spain) due to the introduction of American red swamp crayfish (*Procambarus clarkii*). Hydrobiologica 506-509: 421-426

Rothlisberger, J.D., Chadderton, W.L., McNulty, J., Lodge, D.M. (2011) Aquatic Invasive Species Transport via Trailered Boats: What Is Being Moved, Who Is Moving It, and What Can Be Done. Fisheries 35: 121-132

Schneider, D.W., Ellis, C.D., Cummings, K.S. (2008) A Transportation Model Assessment of the Risk to Native Mussel Communities from Zebra Mussel Spread. Conservation Biology 12(4): 788-800

Scalici, M., and Gherardi, F. (2007) Structure and dynamics of an invasive population of the red swamp crayfish (Procambarus clarkii) in Mediterranean wetland. Hydrobiologia 583: 309-319.

Souty-Grosset, C., Anastácio, P.M., Aquiloni, L., Banha, F., Choquer, J., Chucholl, C., Tricarico, E. (2016) The red swamp crayfish *Procambarus clarkii* in Europe: Impacts on aquatic ecosystems and human well-being. Limnologica – Ecology and Management of Inland Waters 58: 78-93.

Statzner, B., Fièvet, E., Champagne, J., Morel, R., Herouin, E. (2000) Crayfish as geomorphic agents and ecosystem engineers: Biological behavior affects sand and gravel erosion in experimental streams. Limnology and Oceanography 45: 1030-1040

Strayer, D.L. & Dudgeon, D. (2010) Freshwater biodiversity conservation: Recent progress and future challenges. Journal of North American Benthological Society 29(1):344-358

Szela, K. and Perry, W.L. (2013) Laboratory Competition Hierarchies between Potentially Invasive Rusty Crayfish (Faxonius rusticus) and Native Crayfishes of Conservation Concern. American Midland Naturalist 169: 345-353

Taylor, C.A, Schuster, G.A., Wylie, D.B. (2015) *Field Guide to Crayfishes of the Midwest. Manual 15*. Champaign, Illinois: Illinois Natural History Survey.

The Forest Preserves of Cook County (2019) Skokie Lagoons. http://fpdcc.com/skokie-lagoons/

United States Army Corps of Engineers 2014. Great Lakes and Interbasin Mississippi Study (GLMRIS).

United States Army Corps of Engineers, Engineer Research and Development Center, Defense Research, and Engineering Center. Great Lakes Arial Imagery. https://greatlakes.erdc.dren.mil

United States Army Corps of Engineers, Chicago District. Calumet Harbor and River/Chicago Area Waterways System Feasibility Study

United States Army Corps of Engineers, Detroit District (2019) Great Lakes Navigation System Fact Sheets

United States Census Bureau. Chicago Population Estimates, 2018. https://www.census.gov/quickfacts/chicagocityillinois

United States Environmental Protection Agency (USEPA) Facts and Figures about the Great Lakes. https://www.epa.gov/greatlakes/facts-and-figures-about-great-lakes

United States Environmental Protection Agency, Mississippi River/Gulf of Mexico Hypoxia Task Force (USEPA). The Mississippi/Atchafalaya River Basin (MARB). https://www.epa.gov/ms-htf/mississippiatchafalaya-river-basin-marb

Walker, K. F., Thoms, M.C., Sheldon, M.C. (1992) Effects of weirs on the littoral environment of the River Murray, South Australia. Pages 271–292 *in* P. J. Boon, P. Calow and G. E. Petts (eds.), River conservation and management. John Wiley & Sons, Chichester, UK.

Weber, M.J. and Brown, M.L. (2011) Relationships among invasive common carp, native fishes and physicochemical characteristics in upper Midwest (USA) lakes. Ecology of Freshwater Fish 20: 270-278

Weis, J.S. (2010) The role of behavior in the success of invasive crustaceans. Marine and Freshwater Behaviour and Physiology 43(2): 83-98.

Wisconsin Division of Natural Resources (WIDNR). Aquatic Invasive Species, Red Swamp Crayfish.https://dnr.wi.gov/lakes/invasives/AISLists.aspx?species=RED_SWAMP_CRAYF

Wilson, K.A., Magnuson, J.J., Lodge, D.M., Hill, A.M., Kratz, T.K., Perry, W.L., Willis, T.V. (2004) A long term rusty crayfish (*Faxonius rusticus)* invasion: dispersal patterns and community change in a north temperate lake. Canadian Journal of Aquatic Science 61: 2255-2266

Wilson, K.A., Howell, E.T., & Jackson, D.A. (2006). Replacement of Zebra Mussels by Quagga Mussels in the Canadian Nearshore of Lake Ontario: the Importance of Substrate, Round Goby Abundance, and Upwelling Frequency. *Journal of Great Lakes Research*, 32(1), 11-28.

Wilson, C. and Weng, Q. (2010). Assessing Surface Water Quality and Its Relation with Urban Land Cover Changes in the Lake Calumet Area, Greater Chicago. Environmental Management 45(5): 1096-1111

Yue, G.H., Li, J., Bai, Z., Wang, C.M., Feng, F. (2010) Genetic diversity and population structure of the invasive alien red swamp crayfish. Biological Invasions 12: 2697-2706

VITA

Erin O'Shaughnessey was born and raised in Mason, Ohio. She graduated from The Ohio State University in May 2016 with a Bachelor of Science in Environmental Science and a specialization in Water Science. O'Shaughnessey started the Masters of Science program at Loyola University Chicago in June 2016. In January 2018, she was awarded an Advanced Scientific Dive Training Grant from the Women's Diver Hall of Fame.